AR201-13757

"Dr. Kevin N. Baer" <pybaer@ulm.edu> on 05/22/2002 02:25:31 PM

To: Rtk Chem/DC/USEPA/US@EPA
cc: ddixon@deltechcorp.com
Subject: Toluene, p-ethyl- test plan and robust summary

The test plan and robust summary for Toluene, p-ethyl- (CAS #622968) are attached. The sponsor is Deltech Corporation (Sponsor ID). Please confirm receipt of this test plan. Thank you.
Kevin N. Baer, Ph.D.

Test plan.p TOVivo_031920020807 PCMelt_031920020807 PCVapor_03192002080
TOAcute_031920020807 TODevel_031920020807 TORepeat_03192002080
TORepro_031920020807 TOVitro_031920020807 PCBoil_031920020807

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AR201-13757A

EPA'S HPV CHALLENGE PROGRAM: TIER I SCREENING
SIDS DOSSIER FOR P-ETHYLTOLUENE
CAS NO. 622-96-8

DELTECH CORPORATION
BATON ROUGE, LOUISIANA

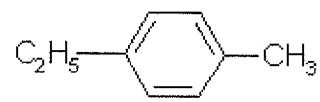
PREPARED BY:
Kevin N. Baer, Ph.D.
The University of Louisiana at Monroe

March 21, 2002

CONTENTS

SIDS PROFILE
HPV Test Plan: Part A

DATE: March 21, 2002

1.01A	CAS NO.	622-96-8
1.01C	CHEMICAL NAME	P-ETHYLTOLUENE
1.01D	CAS DESCRIPTOR	Not applicable
1.01G	STRUCTURE AND FORMULA	 C_9H_{12}

TEST PLAN JUSTIFICATION/ ISSUES FOR DISCUSSION	PHYSICAL/CHEMICAL PROPERTY TESTS DATA GAPS: SIDS testing required: Water solubility, and partition coefficient. ENVIRONMENTAL FATE AND PATHWAY TESTS DATA GAPS: SIDS testing required: Photodegradation, biodegradation, stability in water, and transport (EQC Level III Fugacity Model). ECOTOXICITY TESTS DATA GAPS: SIDS testing required: Acute toxicity to fish, acute toxicity to aquatic invertebrates, and acute toxicity to algae. HEALTH EFFECTS TESTS DATA GAPS: Toxicity to Reproduction; recommend submission of Oral, 2-Generation, Rat study with p-methylstyrene as analogue study for PET. Justification is discussed below.

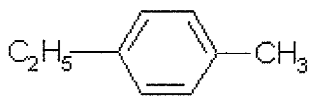
Tier I

DATE: January 15, 2002

HPV Test Plan: Part B							
CAS No:	InfoAvail?	GLP	OECD Study	Other Study	Estim. Meth.	Acceptable?	SIDS Testing Required?
	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N
Physicochemical							
Melting Point	Y	*	*			*	N
Boiling Point	Y	*	*			*	N
Density[1]	Y	*	*			*	N
Vapor Pressure	Y	*	*	--	--	*	N
Oct: water part.coef	N						Y
Water solubility	N						Y
pKa							
Other		-	--	--	--	--	--
Environmental Fate and Pathway							
Photodeg	N						Y
Stability in water	N						Y
Monit. Data[1]	N	--	--	--	--	--	
Transp/Dist	N						Y
Biodeg	N						Y
Other		--	--	--	--	--	--
Ecotoxicology							
Acute Fish	N						Y
Acute Daph.	N						Y
Acute Algae	N						Y
Chron. Daph[2]	N						
Terr. Tox.[2]	N						
Other		--	--	--	--	--	--
Toxicology							
Acute	Y	*	N			*	N
Rep.	Y	Y	N			Y	N
DoseGenetic	Y	Y	N			Y	N
Repro	N			Y	--		N
Devel/Terat	Y	Y	N			Y	N
Human Experience[2]	N						
Other		--	--	--	--	--	--
* Unknown [1] Not required for SIDS Base Set [2] Conditional SIDS studies							

1.0 GENERAL INFORMATION

A.	CAS NUMBER	622-96-8
B.	Molecular Weight	120.194
C.	OECD Name	p-ethyltoluene
D.	CAS Descriptor	Not applicable
E.	Structural Formula	C_9H_{12}



2.0 PHYSICAL/CHEMICAL DATA

2.1 Melting Point

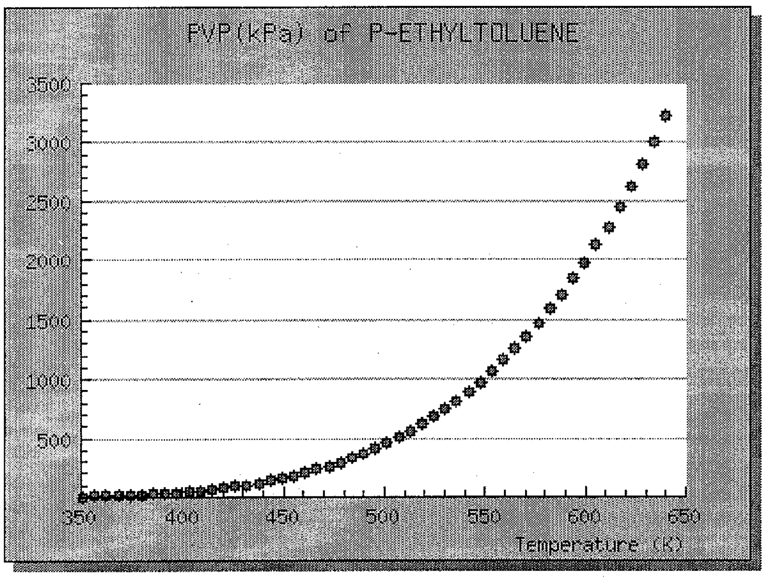
Value:	-62°C
Decomposition	No Data
Sublimation	No Data
Method	No Data
GLP	Yes[] No[] ?[X]
Remarks:	None
Reliability:	[4] Not assignable because limited study information was available
Reference:	Acros Organics (MSDS)

2.2 Boiling Point

Value:	162°C at 760 mm Hg
Decomposition	No Data
Method	No Data
GLP	Yes[] No[] ?[X]
Remarks:	None
Reliability:	[4] Not assignable because limited study information was available
Reference:	Acros Organics (MSDS)

2.3	Water Solubility	No data available Remarks: No testing is needed if water solubility values are ≤ 1 µg/L.
2.4	Vapor Pressure	28 mm Hg at 150°F Remarks: Coefficients available

PVP] Vapor pressure of P-ETHYLTOLUENE



Equation Name	Wagner Equation	
Equation	ln (Pvp/Pc) = (A*x + B*x^1.5 + C*T^3 + D*T^6) / (1-x) where x = 1-T/Tc	
Coefficient A	-7.68892E+00	
Coefficient B	1.92605E+00	
Coefficient C	-5.51788E+00	
Coefficient D	2.76399E+00	
Coefficient E		
Coefficient F		
Coefficient G		
T range , from	351	K
T range , to	640	K

2.5 Partition Coefficient No data available
Remarks: Calculated or estimated values are acceptable.

3.0 ENVIRONMENTAL FATE (Inadequate information)

4.0 ECOTOXICITY (Inadequate Information)

5.0 HEALTH EFFECTS TESTS

5.1 Acute Toxicity (The following tests are available but have not been reviewed)

Oral LD_{50}, Rat	4850 mg/kg
Acute Dermal Toxicity	Moderate skin irritant
Eye Irritation	Moderate eye irritant
Skin Irritation	Moderate skin irritant
LC_{50}, Rat	>3900 ppm
14-Day Dermal	No deaths; severe local skin toxicity

5.2 Repeated Dose Toxicity

5.21 13-Week Oral, Rat

Species:	Fischer 344 rats
Value:	No effects at 100 mg/kg
Method:	Repeated oral gavage doses at 100, 300, and 900 mg/kg daily for 13 consecutive weeks to male and female rats.
Test Substance:	p-ethyltoluene
GLP	Yes[]No[]?[X]
Remarks:	Dose-related mortality, body-weight depression, increase in liver weights, and increases in SGPT, ALP and albumin levels were observed for the 300 and 900 mg/kg males and females. Dose-related reductions in testes/epididymides weights of 300 and 900 mg/kg males were observed.
Reliability:	[2] valid with restrictions
Reference:	MEHSL Sample No. 701-81, Borriston Labs, Inc. 1983

5.22 13-Week Inhalation No effect at 305 ppm; minimal effects at 979 ppm (Not reviewed)

5.23 Range-Finding, Oral Rat (Not reviewed)

5.3 Toxicity to Reproduction (Inadequate Information; p-methylstyrene will be used as an analogue study, see attached justification below).

5.4 Developmental Toxicity/Teratogenicity

5.41 Teratology, Rat

Species:	Pregnant Charles River COBS®CD® rats
Value:	Treatment with PET did not produce a teratogenic response when administered orally to pregnant rats at a dosage level of 200 mg/kg/day or less.
Method:	Oral gavage doses at 25, 100, and 200 mg/kg were administered as a single daily dose on days 6 through 19 of gestation.
Test Substance:	p-ethyltoluene in corn oil (0.5 ml/kg)
GLP	Yes[]No[]?[X]
Remarks:	Survival was 100% in all dosage groups. There were no biologically meaningful or statistically significant differences in any endpoint; mean numbers of corpora lutea, total implantations, early resorptions, postimplantation loss, viable fetuses, or number of litters with malformations.
Reliability:	[2] valid with restrictions
Reference:	(M-3040-79), International Research and Development Corporation, 1981.

5.42 Teratology, Rabbit

Species:	Pregnant Dutch Belted rabbits
Value:	PET did not produce a teratogenic response when administered orally to pregnant rabbits at a dose level of 200 mg/kg/day or less.
Method:	Oral gavage doses at 25, 125, and 200 mg/kg/day were administered as a single daily dose on days 6 through 27 of gestation.
Test Substance:	p-ethyltoluene in corn oil (0.5 ml/kg)
GLP	Yes[]No[]?[X]
Remarks:	No effect related to treatment on Cesarean section parameters or the number of fetuses with malformations occurred in 25, 125, and 200 mg/kg/day. There was an increase in the occurrence of one genetic and developmental variation (13^{th} rudimentary ribs) in the 200 mg/kg/day group. However, 13^{th} rudimentary ribs are considered a skeletal variant and not a malformation.
Reliability:	[2] valid with restrictions
Reference:	(M-3050-79), International Research and Development Corporation, 1981.

5.43 Pilot Teratology, Rat (Not reviewed)

5.44 Pilot Teratology, Rabbit (Not reviewed)

5.45 Pilot Teratology, Rabbit (Not reviewed)

5.5 Genetic Toxicity – *In Vitro*

5.51 Sister Chromatid Exchange Analysis

Species:	Male mouse bone marrow cells
Route of Admin.	Oral gavage
Doses:	0.75, 1.0, and 1.25 g/kg
Test Substance:	p-ethyltoluene suspended in Methocel K4M Premium
Methods:	Bone marrow cells were collected 24 hours after dosing and examined microscopically for sister chromatid exchange.
Results:	PET did not significantly increase the number of sister chromatid exchanges above the baseline vehicle controls.
GLP	Yes[]No[]?[X]
Remarks:	PET does not induce SCE in this test system.
Reliability	[2] valid with restrictions
Reference:	(731-82), Mobile Environmental and Health Science Laboratory, 1983.

5.52 Unscheduled DNA Synthesis

Species:	Male Sprague-Dawley rat hepatocytes
Route of Admin.	Oral gavage
Doses:	0.5, 0.75, 1.0, 1.25, and 1.7 g/kg
Test Substance:	p-ethyltoluene
Methods:	Hepatocytes were isolated two hours after dosing and exposed in culture to ^{3}H-thymidine.
Results:	A significant overall increase in UDS was evident in all assays at doses up to 1.0 g/kg. At higher doses, UDS was diminished, possibly as a function of cytotoxicity.
GLP	Yes[]No[]?[X]
Remarks:	PET is capable of causing primary DNA damage in this test system.
Reliability	[2] valid with restrictions
Reference:	(732-82), Mobile Environmental and Health Science Laboratory, 1983.

5.53 Mitotic Recombination

Specie/Strain:	*Saccharomyces cerevisiae* D_5
Doses:	0.020 to 0.312 µl per 3 ml
Test Substance:	p-ethyltoluene
Methods:	PET was tested for the induction of mitotic recombination in yeast with and without metabolic activation from a rat liver S-9 mixture.
Results:	PET did not induce mitotic recombinations in any of the assays conducted.
GLP	Yes[]No[]?[X]
Remarks:	PET is considered genetically inactive to the indicator strain *Saccharomyces cerevisiae* strain D_5.
Reliability	[2] valid with restrictions
Reference:	(733-82), Litton Bionetics, 1982.

The following tests are available but have not been reviewed:

In Vitro

Ames Assay	Not mutagenic
Mouse Lymphoma	Not mutagenic
DNA Repair	Negative
Cell Transformation	Negative

In Vivo

Drosophila Mutagenicity	Not mutagenic
Dominant Lethal Assay	A statistically significant increase in preimplantation loss was observed in litters sired by male mice given 1300 mg/kg PET in corn oil orally for five days. Genetically induced preimplantation loss cannot be distinguished from failure of fertilization. No increase in fetal death or embryonic resorption occurred at any dose of PET.

Summary of mutagenicity of PET: Although PET demonstrated interaction with DNA in rat hepatocytes and increased preimplantation loss was observed in the dominant lethal test, neither occurrence constitutes a mutagenic event. All other tests which evaluated PET for mutagenesis or cell transformation were negative. There is no clear-cut demonstration of mutagenic activity for PET (Final Status Report, p-methylstyrene, January, 1989).

JUSTIFICATION FOR USING THE STUDY ENTITLED "REPRODUCTIVE EFFECTS OF P-METHYLSTYRENE ADMINISTERED ORALLY VIA GAVAGE TO RATS FOR TWO GENERATIONS" AS AN ANALOGUE REPRODUCTIVE STUDY FOR P-ETHYLTOLUENE

A review was conducted of the available acute and chronic toxicity data and relevant developmental/teratogenicity data for p-methylstyrene and p-ethyltoluene. In my opinion, there are no biological meaningful differences between any endpoints. A brief comparison follows:

p-methylstyrene

Oral LD50 Rat	2523 mg/kg
Inhalation LC50	>3500 ppm
13-week Oral	No effects at 100 mg/kg
13-week Inhalation	No effects at 500 ppm
2-Generation, Rat	No reproductive effects at 200 mg/kg
Teratology, Rat	No effects at 600 mg/kg/day or less
Teratology, Rabbit	No effects at 150 mg/kg/day or less

p-ethyltoluene

Oral LD50 Rat	4850 mg/kg
Inhalation LC50	>3900 ppm
13-week Oral	No effects at 100 mg/kg
13-week Inhalation	No effect at 305 ppm; minimal effects at 979 ppm
Teratology, Rat	No effect at 200 mg/kg/day (highest dose)
Teratology, Rabbit	No effect at 200 mg/kg/day

The OECD SIDS program accepts an existing, adequate 90-day repeat dose study that "demonstrates no effects on reproductive organs, in particular the testes, then a developmental study can be considered as an adequate test for information on reproduction/development effect" (SIDS Manual, Section 3.3, paragraph 13). In a 90-day oral study with Fischer 344 rats, PET was administered at 100, 300, and 900 mg/kg/day for 13 consecutive weeks. Dose-related reduction in absolute and relative testes/epididymides weights of the mid and high dose males were observed. Microscopically, these rats had testicular atrophy and hypospermatogenesis of the testes and hypospermia or aspermia of the epididymides in the high dose males; a number of these animals had sperm granulomas in the epididymides. No microscopic indication of atrophy was seen in the testicles from at 300 mg/kg/day rats; however, two of the animals showed minimal hypospermatogenesis. Testicle sections from all low dose male rats appeared normal. There were no effects at 100 mg/kg/day.

In a teratology study with pregnant rats, PET was administered orally at dose levels of 25, 100, and 200 mg/kg/day. There were no biologically meaningful or statistically significant differences in the mean numbers of corpora lutea, total implantations, early resorptions, postimplantation loss, viable fetuses, the fetal sex distribution, mean fetal body weight, or the number of litters with malformations in any treatment group. Therefore, the conclusion of this study is that treatment with PET did not produce a teratogenic response to pregnant rats at a dose level of 200 mg/kg/day.

These two studies provide adequate test information on reproduction/developmental effects for PET.

Furthermore, comparing the relevant PET endpoints to p-methylstyrene gives relevance in using the p-methylstyrene reproductive toxicity study as an analogue study for PET. For example, a 13-week oral study in rats was conducted with p-methylstyrene using dose levels of 50, 100, 300, 700, and 1500 mg/kg/day. Increases in liver weights were observed at 300 mg/kg/day and decreases in testes weights were observed at 700 mg/kg/day. No significant effects were observed at 100 mg/kg/day.

In the teratology study with pregnant rats, p-methylstyrene was administered orally at dose levels of 60, 190, and 600 mg/kg/day. There were no significant differences in pregnancy, implantation, number of live fetuses, numbers of dead fetuses, or numbers of resorptions per dam between any test level and control. The NOEAL was greater than 600 mg/kg/day.

In view of these considerations, the reproduction study using p-methylstyrene should suffice as an analogue study for PET. Dose levels of 25, 200, 500, and 600 mg/kg/day p-methylstyrene were administered by oral gavage daily for 404 days. There were no effects on the viability of pups from dams dosed at 25 or 200 mg/kg/day. In addition, there was no effect on mating, fertility, gestation, delivery of pups, or lactation index at these dose levels. Therefore the NOAEL and LOAEL were 200 and 500 mg/kg/day, respectively.

Previously, similar comparisons were conducted between p-methylstyrene, vinyl toluene, and styrene. The same conclusions were made; no meaningful differences were apparent between studies. The main metabolites of the isomers of methylstyrene are similar to the corresponding styrene metabolites. There is no indication that metabolites of PET would be different from these related compounds. Therefore, the use of a p-methylstyrene reproductive study as an analogue study for PET is appropriate. The reproductive study for p-methylstyrene was reviewed as part of the Tier II EPA Robust Summary and was determined to be acceptable.

AR201-13757B

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","TestT
ype","GLP","Year","Species","Strain","Sex","NumberofMales","Number
ofFemales","Route","Doses","ExposPeriod","StatMeth","MethodRem","E
ffonMitoticIdx","GenotoxicEff","StatResults","ResultsRem","Conclud
ingRem","Reliability","ReliRem","GeneralRem","RefRem","Completed"
15022002093307.0,1,2/21/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-26-79
Purity: 95.6%
Additions: None reported
Solvent Carrier: None
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.5275","Drosophila
SLRL test","Yes",1979,"Drosophila melanogaster","Not applicable",,
0,0,"Inhalation","0.05 ml vaporized into a half-pint bottle","30-4
0 minutes depending on endpoint","Poisson distribution","The test
substance was evaluated in a battery of standard Drosophila melano
gaster mutagenesis assays. Tests for point mutations included ind
uction of sex-linked lethals and somatic reversion of the white-iv
ory eye color mutant demonstrated by an increase in the frequency
of male flies with red mosaic spots in their eyes. Chromosome abe
rrations and loss were measured by induction of dominant lethal mu
tations, Y chromosome loss and the bithorax test of Lewis in which
chromosome rearrangements stimulate the development of a band of
hairy tissue between the thorax and abdomen, the metanotum.

The treatment consisted of 0.05 ml of the test substance on filter
paper placed within half-pint bottles. Adult flies were exposed
for 30 minutes and larvae were exposed for 40 minutes. Mitomycin
C (40 ug/ml, 1 hr) was used as the positive control in the somatic
reversion tests, ethylmethanesulfonate (EMS, 0.04 M) was used in
the induction of dominant lethal mutations and sex-linked lethals
tests, and x-rays (3 krads) were used in the Y-chromosome loss and
bithorax test of Lewis studies.

In the timing of crosses, four days are required for the maturatio
n of the Drosophila sperm cell after meiosis. In all crosses, tre
ated males were mated for three days only to assure use of a very
uniform population of treated cells and to avoid confounding the d
ata by the occurrence of clusters of mutations arising from the di
vision of a mutated stem cell.

Since the events scored in these tests have very low probabilities
, their analysis is best based on the poisson distribution. Fiduc
ial limits are computed according to Stevens.","Not applicable","N
egative","All endpoints were not significantly different from cont
rol at p <0.05.","Initial studies showed the test substance to be
extremely toxic to Drosophila. Exposure to 0.05 ml vaporized into
a half-pint bottle containing the flies anesthetized all the flie

s within 30 minutes, killed about one-third, and did not sterilize the survivors. Larvae responded similarly to a 40-minute treatment.

Results of the somatic reversion of the white-ivory eye color mutant test is as follows (number scored, number of mosaics, frequency of mosaics, respectively): Control, 1096, 5, 0.005; MCTR-26-79, 2126, 8, 0.004; Positive control (mitomycin C), 108, 17, 0.16. The test substance is not significantly different from control. With 95% probability, the true mosaic frequency following treatment does not exceed 0.007.

Results of the sex-linked lethal tests (number scored, number of lethals, frequency of lethals, respectively): Control, 6760, 7, 0.001; MCTR-26-79, 2197, 3, 0.001; Positive control (EMS), 69, 16, 0.23. The test substance is not significantly different from controls. With 95% probability, the true frequency of lethal mutations following treatment does not exceed 0.0035.

Results of the induction of dominant lethal mutations test are as follows (number scored, number inflated, frequency of lethals, respectively): control, 1025, 18, 0.018; MCTR-26-79, 2402, 29, 0.012; Positive control (EMS), 247, 8, 0.03. The test substance is not significantly different than controls. With 95% probability, the true frequency of inflated eggs after treatment does not exceed 0.016.

Results of the Y chromosome loss test (total number, number X/0, frequency of loss, respectively): control, 1084, 3, 0.003; MCTR-26-79, 2048, 4, 0.002; Positive control (x-rays), 449, 13, 0.03. The test substance is not significantly different from controls. With 95% probability, the true frequency of chromosome loss following treatment does not exceed 0.0045.

Results of the bithorax test of Lewis (number tested, transvections, frequency, respectively): control, 1322, 0, 0; MCTR-26-79, 2140, 0, 0; Positive control (x-rays), 110, 6, 0.06. The test substance is not significantly different from controls. With 95% probability, the true frequency of transvections following treatment does not exceed 0.0014.","Treatment with MCTR-26-79 did not signficantly increase the frequency of mutagenic endpoints over control values in any of the assays. It appears that this test subtance does not induce genetic damage in Drosophila melanogaster under the experimental conditions used.","Acceptable","The key parameters (i.e., dose preparation, exposure times, use of positive controls) were appropriate and adequately described in the study.",,"Drosophila Mutagenicity Assays of Mobil Chemical Company Compound MCTR-26-79. Study No.: 009-617-278-9. EG&G Mason Research Institute,Rockvil

le, Maryland, September 28, 1979 (M262-79).","N"
15022002093307.0,2,2/28/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: 01038003
Purity: Assume 100% for dosing calculations (specific gravity, 0.8
6 g/ml)
Additions: None reported
Solvent Carrier: Methocel K4M (10 ml/kg)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.5915","Sister chro
matid exchange assay","Yes",1983,"mouse","Swiss Webster","M",5,0,"
Oral","0.75,1.00, and 1.25 gm/kg","single exposure","Student's t-t
est analysis","The purpose of this study was to investigate whethe
r the test substance is capable of causing a significant increase
in sister chromatid exchanges in the bone marrow of mice treated i
n vivo. Twenty-five male Swiss Webster mice (approximately 25 gra
ms) were dosed by oral gavage with the test substance at 0.75, 1.0
0, and 1.25 g/kg. A positive control (cyclophosphamide, 7.5 mg/kg
) was given by intraperitoneal injection. The vehicle (negative c
ontrol) was Methocel K4M and was given by oral gavage (10 ml/kg).
Five animals were used for each test group. All animals were dos
ed on consecutive days in one week with at least one animal from e
ach dose group dosed per day.

5-Bromodeoxyuridine (BrdU) was pressed into a 50 mg tablet and imp
lanted subcutaneously in the skin-fold at the back of the neck. T
he wound was clamped with surgical wound clips. Six hours after B
rdU implantation, the animals were dosed with the test substance.
Colchicine (1 mg/ml) was given IP (2 mg/kg), to arrest cells in m
itosis, twenty hours after dosing.

Two hours after colchicine treatment, the animals were killed and
both femurs were processed. A minimum of five slides were made fo
r each animal and differentially stained. Twenty-five cells/anima
l were scored when possible. One hundred cells were classed as be
ing in metaphase 1, 2, or 3 after BrdU exposure to determine if th
e test substance caused a cell cycle delay. Metaphase cells which
have replicated once (metaphase 1) have chromatids that are stain
ed with equal intensity by Hoechst stain. Those which have replic
ated twice (metaphase 2) have a chromatid, that is bifilarly subst
ituted with BrdU, staining lighter, and a unifilarly substituted c
hromatid staining darker. Metaphase cells that have replicated th
ree times (metaphase 3) have two bifilarly substituted chromatids.

Student's t-test statistics were performed to compare test values
with the vehicle control. A significant increase in the occurrenc
e of SCEs above background levels is an indication of potential mu
tagenic activity by the test substance.","No detectable cell cycle
delay was observed.","Negative","No significant increases in SCEs

between treatment groups and vechicle control at p<0.001. The positive control was highly significant from control at p<0.001.","The test substance did not induce any remarkable pharmacologic effects. The animals treated with the test substance did not have a significant increase in SCEs above the vehicle control-treated animals according to Student's t-test, nor was there a detectable cell cycle delay observed. Cyclophosphamide was found to induce a highly signficant increase in SCE levels.

The average SCE/Cell (based on 40 chromosomes per spread) for vehicle control, test substance (0.75, 1.00, and 1.25 g/kg), and positive control was 7.45, 9.98, 8.32, 6.91, and 16.65, respectively. The average SCE/Chromosome was 0.19, 0.25, 0.21, 0.18, and 0.42, respectively. The average number of Chromosomes/Cell was 39.7, 39.4, 39.4, 39.4, and 39.6, respectively.","The test substance did not induce statistically signficant increases in sister chromatid exchanges in bone marrow cells from the treated mice and therefore, is not considered a DNA damaging agent under these test conditions.","Acceptable","The key parameters (i.e., number of doses, positive controls, etc.) were appropriate and adequately described in the study.",,"Sister Chromatid Exchange Analysis of Mouse Bone Marrow Cells Treated In Vivo with Para-Ethyltoluene. Study No.: 20731. Mobile Environmental and Health Science Laboratory, Plainsboro, New Jersey, June 2, 1983 (731-82).","Y"
15022002093307.0,3,3/3/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: PET
Purity: Assume 100% for dosing calculations specific gravity, 0.89 g/ml)
Additions: None reported
Solvent Carrier: Methocel K4M Premium (0.75%; 10 ml/kg))
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.5550","Unscheduled DNA synthesis","Yes",1983,"rat","Sprague-Dawley","M",16,0,"Oral","0.50, 0.75, 1.00, 1.25, and 1.70 g/kg","Single oral dose.","Chi square analysis and Spearman Correlation tests","The purpose of this study was to investigate the potential of the test substance to cause primary DNA damage by measuring unscheduled DNA synthesis (UDS) in a primary culture of rat hepatocytes treated in situ. Sixteen male Sprague-Dawley rats (averaging 184 g) were used. Animals were given a single oral dose of the test substance suspended in Methocel K4M Premium (10 ml/kg). Two UDS experiments were conducted; the first experiment used doses of 0.50, 1.00, and 1.70 g/kg and the second experiment used doses of 0.75, 1.00, and 1.25 g/kg. The positive control (2-acetylaminofluorene, 2-AAF, 20 mg/kg) and negative controls (Methocel K4M, 10 ml/kg) were also given orally in one administration. There were four animals assigned to each test substance group and two assi
gned for the positive and two for the negative solvent control gro

ups. Two hours after dosing the hepatocytes were isolated by liver perfusion.

Hepatocyte isolation was conducted as follows: rats were sacrificed by cervical dislocation 2 hours after dosing. Livers were perfused in situ (inferior vena cava cannulated and the portal vein sectioned) for 5 minutes with 0.5 mM ethyleneglycol bis (beta-aminoethyl ether)-N, N'-tetraacetic acid (EGTA) in calcium and magnesium free Hanks' Balanced Salt Solution buffered with 0.05 M HEPES solution at pH 7.2. A 10-minute enzyme perfusion was performed with Type I collagenase solution made to 0.15 mg/ml in Williams' Medium E. The livers were minced in fresh collagenase solution at room temperature and filtered into a centrifuge tube. Complete Williams' Medium E, containing 10% Fetal Bovine Serum, 10 milliunits/ml insulin, 1 uM dexamethasone, and 50 ug/ml Garamycin, was added to the suspension of primary hepatocytes to arrest collagenase activity. The cell suspension was centrifuged at 50 x g for 4 minutes.

Five tissue culture plates (60-mm) were inoculated with 3 ml of complete medium containing 1E+6 viable cells/ml and incubated for 2-4 hours at 37 degrees C with 5% carbon dioxide in air. Unattached cells were removed and 3 ml of fresh media containing 10 uCi/ml of 3H-thymidine were added to each plate. Cells were incubated for 18 hours at 37 degrees C in 5% carbon dioxide in air.

Following 18 hours incubation with 3H-thymidine, the cultures were washed three times. Cells from each liver were removed from the plates and pooled in a 0.001 M phosphate buffer with 0.32 M sucrose, 0.0015 M calcium chloride and 1% Triton X-100. The cells were homogenized in the buffer with a Wheaton Dounce Homogenizer to release nuclei. Nuclei were centrifuged at 1000 rpm (200 x g) for 5 minutes, resuspended in fresh buffer, and fixed in 3:1 fixative (methanol:acetic acid) at 4 degrees C. A minimum of five slides were made from the pooled cultures derived from each rat.

Twenty-four hours after the slides were made, they were dipped in Kodak NTB-2 autoradiographic emulsion and dried. The coated slides were stored for 2 weeks at 4 degrees C in light-tight containers with Drierite. The emulsions were developed in D-19, fixed and stained with 3% Giemsa.

All slides were coded and 1000 cells per animal were scored and assigned to the following categories: background (0-6 grains), >background but <50 grains, or S phase (>50 grains). Chi Square analysis was performed to compare test values to negative control data; a significant increase in UDS is an indication of primary DNA damage and repair activity induced by the test substance. Statistical Analysis Systems (SAS) computer assisted analysis was used to tes t dose relationships; the program used Spearman non-parametric ana lysis for comparisons.

A third experiment was conducted to determine whether the test sub stance has the ability to stimulate scheduled DNA synthesis (S-pha se) which in early stages may be mistaken for UDS. The procedures and doses for experiment #2 were followed. Parallel cultures (fo ur plates with Hydroxyurea, HU, an agent that inhibits S-phase and four without) were established for each test liver and the 2-AAF dose was increased to 25 mg/kg. The duplicate cultures contained HU. The slides from this experiment were scored by counting silve r grains/nucleus to obtain a mean count of grains/nucleus, rather than the categorization method described earlier. In an experimen t to ascertain the induction of S-phase, grains/nucleus is a more sensitive means of measurement. Student's t-test was used to eval uate the data.

The animals used for analytical samples were not the animals used for UDS experimentation. These experiments were conducted indepen dently of the genetic perturbation assays. Headspace analysis of blood from rats dosed with the test substance was performed by gas chromatograph/mass spectrophotometer (GC/MS) equipped with 30 met er x 0.25 mm (i.d.) DB-5 fused silica capillary column. Samples o f whole blood were extracted with hexane and analyzed by GC equipp ed with a flame ionization detector (FID). Perfused liver samples were also analyzed by GC/FID. A 1 g sample was homogenized with 0.5 ml hexane for 1 minute. The homogenate was then centrifuged a t 4000 rpm for 20 minutes and the extract was analyzed for the tes t substance by GC/FID.","Not specifically measured.","Positive","A significant (Chi Square; <0.05) overall increase in UDS was evide nt in all assays at doses up to 1.0 g/kg. Spearman Correlation te sts found a positive correlation between dose and UDS response."," Primary hepatocyte cultures from s
everal animals did not meet test criteria. Since all of the slide s from the study were coded and evaluated, data from these culture s were collected but were not included in final tabulations.

Analytical data indicate that PET was absorbed into the blood and was also detected in the liver at the same time of cell harvest. For example, 1.7 g/kg at 2, 4, and 6 hrs resulted in mean blood le vels of 29.5, 39.7, and 35.6 ppm, respectively. For 2.1 g/kg at 2 , 4, and 6 hrs, mean blood levels were 42.3, 27.4, and 42.5 ppm, r espectively. The mean levels of PET in livers for 1.0 g/kg at 2 a nd 4 hrs were 60.0 and 61.3 ppm, respectively. The mean levels of PET in livers for 1.7 g/kg at 2 and 4 hrs were 120 and 54 ppm, re spectively.

A significant overall increase (Chi Square, p<0.05) in UDS was evi

dent in all assays at doses up to 1.0 g/kg. At higher doses, UDS activity was diminished, possibly as a result of cytotoxicity. A cytotoxic effect was evidenced by a reduced ability of the hepatoc ytes from rats given 1.7 g/kg to attach to the petri plates for cu lturing. In addition to the standard Chi Square analysis, the dat a were also analyzed by non-parametric procedures (Spearman Correl ation tests) using four variables; S phase cells, UDS cells, backg round cells and dose. The total data package, including data from animals excluded by test criteria, was tested for correlation bet ween cited variables with and without the high dose data to ascert ain the effect of apparent cytotoxicity on the correlation. The d ata were also analyzed in the same manner with the data from the e xcluded animals deleted from the analyses. With one exception, al l of the corrrelation coefficients as calculated between dose and UDS response went from a negative
correlation to a distinctly positive correlation when the high dos e was dropped from the analysis; the exception (experiment three; without HU) went from 0.03643 to 0.42817, which is a distinct chan ge to a more positive trend. The fact that the trend shift in cor relation is reproducible across different experiments is a strong indication that the statistically significant positive response by Chi Square is valid.

The number of cells classified as undergoing UDS (1000 cells/anima l) for experiment 1 was as follows (for solvent control, positive control, 0.5, 1.0, and 1.7 g/kg, respectively): total cells, 2000, 1000, 4000, 3000, and 3000. UDS, 230, 486, 567, 702, and 209. P ercent UDS, 11.5, 48.6 (significant at $p<0.001$), 14.2 (significant at $p<0.05$), 23.4 (significant at $p<0.001$), and 6.97% (less than s olvent controls due to cytotoxicity, significant at $p<0.001$). The number of cells classified as undergoing UDS (1000 cells/animal) for experiment 2 was as follows (for solvent control, positive con trol, 0.75, 1.0, and 1.25 g/kg, respectively): total cells, 2000, 2000, 2000, 3000, and 4000. UDS, 151, 982, 125, 299, and 174. Pe rcent UDS,7.6, 49.1 (significant at $p<0.001$), 6.2, 10.0 (significa nt at $p<0.001$), and 4.4% (less than solvent controls due to cytoto xicity, significant at $p<0.001$).

It was observed that within a given dose group, cell populations f rom some animals demonstrated increased UDS activity while others had UDS levels equal to or below that of solvent controls. The as say was performed three times over similar dose ranges and despite intergroup variability, the same pattern of response was observed in each test. The third test was specifically designed to determ ine whether the test substance stimulated scheduled (S-phase) DNA synthesis which could, in the early stages of replication, be conf used with UDS because of the low levels of 3H-TdR incorporation. If PET-stimulated S-phase synthesis was responsible for the observ

ed activity, introduction of HU into parallel PET-treated hepatocy te cultures derived from the same rat liver would inhibit both S-p hase synthesis and the spurious UDS activity. Data from experiment 3 demonstrate, although HU did slightly inhibit the spontaneous i ncidence of cells in S-phase in most cultures, the test substance did not reduce the percentage of c
ells undergoing UDS. For example, mean cell viability for experim ents 1, 2, and 3 were 90.1, 98.8, and 87.02%, respectively. The m ean number of cells/liver for experiments 1, 2, and 3 were 3.36E+7 , 3.69E+7, and 2.17E+7, respectively. When the average grains per nucleus were visualized, it was apparent that PET did not stimula te S-phase synthesis and the addition of HU did not significantly alter the pattern of UDS activity. A statistically significant (t -test, $p<0.05$) increase in grains/nucleus was observed at 0.75 but not at 1.0 g/kg . The higher background values for UDS in this ex periment may be due to higher levels of radioactivity of the 3H-Td R or more sensitive developing emulsion. However, the overall pat tern of response is similiar to that reported in the first two stu dies, a signficant increase in UDS over a narrow dose range and th en diminished response at the high dose.","PET induced a significa nt increase in UDS in rat hepatocytes when compared to negative co ntrol values. A dose-dependent in
crease was not observed, possibly because of severe cytotoxicity a t higher doses which inhibited repair of primary DNA damage. Howe ver, the reproducibility of the response indicates that PET produc ed primary DNA damage observable in a narrow dose range as UDS.

It was observed that within a given dose group, cell populations f rom some animals demonstrated increased UDS activity while others had UDS levels equal to or below that of solvent controls. This t ype of animal to animal variability has been observed in other stu dies (Williams, 1977, Cancer Res. 37:1845-1851). Variation may be due to variability in biotransformation between animals. Physical variables of a technical nature such as the age of the tritiated thymidine, emulsion or duration of exposure can attribute to varia bility. To compensate for animal to animal variability, more test animals were assigned to each test group and criteria to eliminat e outlier cell populations were stringent. The occurrence of resp onding and non-responding cell populations within a dose group and activity over a narrow dose range suggest PET may induce two comp eting phenomena in exposed cell populations, mediated in part by v ariation in individual host animal metabolism. The increase in UD S in some animals may be in respon
se to perturbed DNA. UDS levels at or below solvent control value s in other animals at the same dose may be due to inhibition of re pair synthesis or possible delay of general metabolic activity.
At the highest doses, overt cytotoxicity was demonstrated by the f ailure of the cells to attach to the petri plates. This cytotoxic

effect also varied between cultures established from livers treat ed at the same dose level.

Despite variability, a statistically significant increase in UDS w as induced by PET in three separate studies, producing a similiar pattern of repair activity over a narrow dose range. It is expect ed, in this type of assay, that at concentrations greater than the cytotoxic level, the rate of UDS will decrease. Since the inhibi tory effect of cytotoxicity on UDS precludes the extension of the dose response curve, the importance of reproducing a positive effe ct over the small dose range for UDS is critical (Mitchell, et al. , 1983, Mutation Res. 123:363-410). Therefore, the results of thi s study strongly suggest that PET is capable of causing primary DN A damage in this test system.","Acceptable","All key parameters (i .e., doses, use of positive and negative controls, etc.) were appr opriate and adequately described in the study.",,"Detection of Uns cheduled DNA Synthesis in Primary Rat Hepatocytes Treated In Vivo with Para-Ethyltoluene. Study Number: 20732, Mobil Environmental and Health Science Laboratory, Pen
nington, New Jersey, May 6, 1983 (732-82).","Y"
15022002093307.0,4,3/18/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: Sample-01038003 (T1609)
Purity: Assume 100% for dose calculations (actual not provided)
Additions: None reported
Solvent Carrier: Olive oil (1 ml/kg)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.5450","Dominant le thal assay","Yes",1980,"rat","Sprague-Dawley","M",10,0,"Oral","0.1 5, 0.5, and 1.5 ml/kg/day","Once daily for 5 consecutive days","t-test, Chi-square, Analysis of regression","The purpose of this stu dy was to determine the mutagenic potential of the test substance to induce fetal wastage in Sprague-Dawley rats. Male and female r ats were quarantined for 10-14 days. Male rats (8-10 weeks old, 1 0 per treatment) were dosed daily by oral gavage for five consecut ive days with negative control (olive oil, 1 ml/kg), 0.15, 0.5, a nd 1.5 ml/kg/day. A positive control (triethylenemelamine, TEM, s ingle intraperitoneal injection of 0.5 mg/kg on day 4) was also us ed. Three days after the last treatment, each male was mated with two virgin females (8-10 weeks old) over a five day period. The male was then allowed to rest for two days, after which the mating process was repeated with two new virgin females. This process w as repeated until the males had be
en mated for seven weeks with two females per week.

Fourteen days from the mid-point of the mating period, the females were sacrificed by carbon dioxide asphyxiation and the abdominal cavity was exposed. The membrane was removed from each ovary and the corpora lutea for each ovary was counted and recorded separate ly. In addition, both uterine horns were examined and fetal death s and total implantations were determined and recorded separately for each horn.

Nine parameters were analyzed in this study; fertility index, aver age number of implantations per pregnant female, corpora lutea per pregnant female, preimplantation losses per pregnant female, dead implants per pregnant female, proportion of pregnant females with one or more dead implants, proportion of pregnant females with tw o or more dead implants, dead implants per total implants, and liv e implants per pregnant female.

The statistical methods used to analyze the data (if appropriate) included the following: t-test, ch-square analysis, analysis of re gression, analysis of linear trend, analysis of variance, and prob it analysis.

The criteria of determination of a valid test was as follows: fema les mated with negative control males must show a total of 8-15 im plantations and females mated to positive control males must exhib it severe fetal damage. There must be a statistically significant reduction in implantations relative to the negative controls and there must be a statistically significant increase in females with two or more dead implants relative to the negative controls. The damage must be seen between weeks 2 and 7 of the spermatogenic cy cle.","None","Negative","T1609 at 1.5 ml/kg/day exhibited signific antly higher (p<0.05) preimplantation losses than the negative con trol.","Two rats receiving 1.5 ml/kg/day died on the third and fif th day of dosing, respectively. In both cases, necropsy revealed severe, extensive hemorrhage in the small intestine. Weight loss was also noted in those animals receiving 1.5 ml/kg/day. Two addi tional animals were dosed with 1.5 ml/kg/day and replaced the dead rats in the study. No adverse ef
fects were observed in 0.5 or 0.15 ml/kg/day groups. Mean body we ights (grams) for negative control, 0.15, 0.5, and 1.5 ml/kg/day, day 5, were 331, 294, 321, and 329, respectively.

The fertility index was calculated by dividing the number of pregn ancies by the number of mated females. On weeks 5 and 6, T1609 at a dose of 1.5 ml/kg/day significantly reduced (p<0.05) the fertil ity index relative to the negative control. TEM had no adverse ef fects. The fertility index for negative control, positive control , 0.15, 0.5, and 1.5 ml/kg/day, week 5, was 0.85, 0.85, 0.70, 0.75 , and 0.50 (significant at p<0.05), respectively. The fertility i ndex for week 6 was 0.90, 0.90, 0.90, 0.90, and 0.50 (significant at p<0.05), respectively.

Implantations per pregnant female represent the average number of

implantations, live or dead, per pregnant female. On weeks 1, 2, 4, 5, and 7, T1609 at 1.5 ml/kg/day significantly reduced (p<0.05) the implantations per pregnant female relative to the negative co ntrol. The linear regression analysis, analysis of variance acros s all weeks grouped into three stages of spermatogenesis suggest a significant (p<0.05) dose-related effect. TEM markedly reduced t he implants per pregnant female on weeks 1-4. Implantations per p regnant female for negative control, positive control, 0.15, 0.5, and 1.5 mg/kg/day (*indicates significant from negative control at p<0.05), week 1, were 10.9, 0.9*, 8.0, 9.7, and 0.3*, respectivel y. Week 2 was 10.8, 1.0*, 10.6, 8.3, and 2.8*, respectively. Wee k 3 was 10.9, 0.6*, 10.0, 10.6, and 7.2, respectively. Week 4 was 12.3, 2.7*, 8.6, 13.2, and 5.8*, respectively. Week 5 was 11.0, 10.5, 10.3, 9.9, and 5.1*, respectively. Week 6 was 15.0, 14.0, 1 4.8, 14.1, and 13.9, respectively.
Week 7 was 13.5, 12.9, 13.3, 12.8, and 10.3*, respectively.

The corpora lutea per pregnant female was variable. The variabili ty in the numbers reflect individual variation in the female rats rather than biological activity of the doses tested, assuming that the test substance or its metabolites are not transmitted via spe rm to the female rats.

Preimplantation losses per pregnant female were estimated by the d ifference between the number of corpora lutea and total implantati ons. On weeks 1, 2, 4, 5, and 7, T1609, at a dose level of 1.5 ml /kg/day significantley increased (p<0.05) the preimplantation loss es. The linear regression analysis, analysis of variance across a ll weeks, and across weeks grouped into stages of spermatogenesis suggest a significant (p<0.05) dose-related effect. TEM markedly increased preimplantation losses on weeks 1-4. Preimplantation lo sses per pregnant female for negative control, positive control, 0 .15, 0.5, and 1.5 ml/kg/day (*indicates significant from negative control at p<0.05), week 1, was 2.42, 12.0*, 5.56, 4.0, and 12.9*, respectively. Week 2 was 2.69, 11.0*, 3.11, 5.41, and 9.67*, res pectively. Week 3 was 4.31, 13.15*, 4.08, 5.79, and 8.3, respecti vely. Week 4 was 2.53, 10.0*, 4.22, 0.7, and 7.3*, respectively. Week 5 was 1.94, 2.76, 2.86, 3.6, and 9.4*, respectively. Week 6 was 0.28, 1.5, 0.5, 0.44, and 1.2
, respectively. Week 7 was 1.33, 1.0, 0.45, 2.26, and 4.5*, respe ctively.

The average number of dead implants per pregnant female was not st atistically different between any of the treatment groups and nega tive control. TEM markedly increased the proportion of pregnant f emales with one or more dead implants on week 5. The proportion o f pregnant females with two or more dead implants and the average number of dead implants per total implants statistically increased

(p<0.05) at 0.5 ml/kg/day on week 2. Since no significant increa ses were observed at 1.5 ml/kg/day on week 2, these effects were n ot biologically meaningful. TEM markedly increased the proportion of pregnant females with two or more dead implants on weeks 5 and 6, and markedly increased the number of dead implants per total i mplants on weeks 1, 2, 4, and 5. The dead implants per total impl ants for negative control, positive control, 0.15, 0.5, and 1.5 ml /kg/day (*indicates significant from controls at p<0.05), week 1, was 0.04, 1.0*, 0.08, 0.15, and 1.0, respectively. Week 2 was 0.0 2, 0.91*, 0.06, 0.12*, and 0.09, r
espectively. Week 3 was 0.03, 1.0, 0.03, 0.07, and 0.11, respecti vely. Week 4 was 0.02, 0.90*, 0.03, 0.11, and 0.07, respectively. Week 5 was 0.03, 0.35*, 0.08, 0.06, and 0.18, respectively. Wee k 6 was 0.03, 0.09, 0.02, 0.03, and 0.03, respectively. Week 7 wa s 0.03, 0.12, 0.05, 0.05, and 0.04, respectively.

The average number of live implants per pregnant female was signif icantly reduced by T1609 at 1.5 ml/kg/day on weeks 1, 2, 4, 5, and 7. The linear regression analysis, the analysis of variance acro ss all weeks, and across weeks grouped into three stages of sperma togenesis suggest a significant (p<0.05) dose-related effect. TEM markedly reduced the number of live implants per pregnant female on weeks 1-5. The live implants per pregnant female for negative control, positive control, 0.15, 0.5, and 1.5 ml/kg/day (*indicate s significant from negative control at p<0.05), week 1, were 10.5, 0.0*, 7.33, 8.21, and 0.0*, respectively. Week 2 was 10.54, 0.09 *, 9.89, 7.29, and 2.5*, respectively. Week 3 was 10.56, 0.0*, 9. 69, 9.79, and 6.4, respectively. Week 4 was 12.07, 0.27*, 8.33, 1 1.8, and 5.4*, respectively. Week 5 was 10.65, 6.76*, 9.5, 9.27, and 4.2*, respectively. Week 6 was 14.5, 12.78, 14.56, 13.67, and 13.5, respectively. Week 7 was 13.0, 11.35, 12.65, 12.11, and 9. 88*, respectively.","The positive
and negative controls fulfilled the requirements for a valid test.

T1609 at 1.5 ml/kg/day exhibited significantly higher preimplantat ion losses than the negative control. The concomitant reduction i n live implants without an increase in dead implants is presumably due to preimplantation embryonic loss. T1609 at 0.5 ml/kg/day ex hibited a higher frequency of dead implants per pregnant female th an the negative control at weeks 1, 2, and 4, although none were i ndividually significant. Since preimplantation losses cannot be d istinguished from failure of fertilization, which may result from a number of factors, fetal death was used as the primary measure o f dominant lethality. T1609 failed to induce a statistically sign ificant increase in dead implants per pregnancy accompanied by a r eduction in live implants per pregnant female.

No detectable mutagenic activity, as defined by induction of fetal death, was found for the test substance in the dominant lethal as say. However, due to the marked increased in preimplantation loss es the results of this study should be evaluated only in conjuctio n with other in vivo or in vitro tests that monitor genetic activi ty.","Acceptable","All key parameters (i.e., use of negative and p ositive controls, doses, etc.) were appropriate and adequately des cribed in the study.",,"Activity of T1609 in the Dominant Lethal A ssay in Rodents for Mutagenicity. Microbiological Associates, Aug ust, 29, 1980 (832-80).","Y"

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","GLP","Year","MethodRem","Prec","MeltingVal","Upper","Unit","Decomposition","Sublimation","ResultsRem","ConcludingRem","Reliability","ReliRem","GeneralRem","RefRem","Completed"
15022002093307.0,1,3/18/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: Toluene, p-ethyl-
Purity: Assume 100%
Additions: Unknown
Solvent Carrier: Unknown
Contaminants: Unknown
Chemical formula: C9H12",,"Unknown","Unknown",,"Unknown","=",-62,0,"§C",,,"Report not evaluated.","The report was not obtained and evaluated.","Unknown","The report was not evaluated.",,"Standard MSDS.","N"

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","GLP",
"Year","MethodRem","Prec","VapourPresVal","Upper","Unit","Temp","D
ecomposition","ResultsRem","ConcludingRem","Reliability","ReliRem"
,"GeneralRem","RefRem","Completed"
15022002093307.0,1,3/18/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: Toluene, p-ethyl-
Purity: Assume 100%
Additions: Unknown
Solvent Carrier: Unknown
Contaminants: Unknown
Chemical formula: C9H12",,"Unknown","Unknown",,"Unknown","=",28.00
,0.00,"mm Hg","65.5",,"Unknown","Report not evaluated.","Unknown",
"The report was not obtained and evaluated.",,"Standard MSDS for T
oluene, p-ethyl-.","N"

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","GLP",
"Year","Species","Strain","Sex","NumberofMales","NumberofFemales",
"Vehicle","Route","MethodRem","Prec","Value","Unit","DeathsperDose
","ResultsRem","ConcludingRem","Reliability","ReliRem","GeneralRem
","RefRem","Completed"
15022002093307.0,5,3/5/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-51-79
Purity: Assume 100% for dose calculations
Additions: None reported
Solvent Carrier: None
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.1300","Unknown",19
79,"rat","Sprague-Dawley","Both",5,5,"None","Inhalation","Two six-
hour inhalation exposures were performed to determine the acute to
xicity of MCTR-51-79 in rats. For the first exposure, the test su
bstance was placed in a 1,000 ml Erlenmeyer flask (used as a reser
ve) and a 1,000-ml bubbler. The bubbler was heated in a 90 degree
s C waterbath using a magnetic stirrer/hot plate. Nitrogen was pa
ssed through the bubbler at approximately 15 liters per minute to
create a vapor. The vapor-laden air then passed through a kjeldah
l trap tube and through a trap flask before entering the 760-liter
 exposure chamber housing the test animals. Another 500-ml volume
tric flask was attached to the first trap to also trap excess comp
ound. Chamber air flow was maintained at 173 liters per minute th
roughout the exposure. Test substance from the reserve Erlenmeyer
 flask was pumped into the bubbler whenever necessary by a Fluid M
etering Pump. All glass surfaces
(flasks, tubing, etc.) were covered with aluminum foil to prevent
light from reaching the test substance and to keep heat within the
 generating system, thus reducing recondensation of the heated vap
or. The test substance, bubbler, reserved flask, trap flask, volu
metric flask, Kjeldahl trap, tubing, clamps, stoppers, and foil we
re weighed before and after the exposure period. The difference i
n weight represented the amount of test substance consumed during
the exposure. The nominal concentration was calculated by dividin
g the amount of material delivered by the total air flow through t
he chamber during the exposure period. Chamber air concentration
was monitored continuously during the exposure using a Miran IA Am
bient Air Analyzer and recorder once each hour. Waterbath tempera
ture, nitrogen flow rate, and chamber air flow were also recorded
hourly.

For the second exposure, a similar procedure was used. However, t
he nitrogen flow rate was approximately 12.5 liters per minute, th
e bubbler was heated in a 80 degrees C waterbath, and the trap use
d was a 1,000 milliliter three-neck round-bottom flask. A volumet
ric flask to trap excess test substance was not used.

The test animals for both groups consisted of five male and five f
emale Sprague-Dawley rats (body weights ranged from 212 to 267 g f
or the first experiment and 217 to 296 g for the second experiment
). The animals in both experiments were observed prior to exposur
e to ascertain their basic health status. Observations for abnorm
alities were made at 15-minute intervals during the first hour of
the exposure, hourly through the termination of the exposure, upon
 removal from the chamber, hourly for two hours post-exposure, and
 daily thereafter for 14 days. Individual body weights for both e
xperiments were recorded on Day 0 (prior to exposure), Day 1, Day
2, Day 4, Day 7, and Day 14. On Day 14, the animals were sacrific
ed with ethyl ether and gross necropsy examinations were performed
.",">",3900,"ppm(air)","No mortality was observed at either concen
tration.","No mortality was observed in any exposure. The analysi
s of chamber concentrations in Experiment 1 was as follows: minute
s into exposure; 70, 120, 179, 243
, 302, and 358 resulted in a chamber concentration of 4250, 3690,
3580, 3580, 4040, and 4250 ppm, respectively. The mean chamber co
ncentration for Experiment 1 was 3900 ppm. The analysis of chambe
r concentrations in Experiment 2 was as follows: minutes into expo
sure; 60, 120, 178, 240, 298, and 354 resulted in a chamber concen
tration of 1740, 2110, 2090, 1920, 2000, and 1900 ppm, respectivel
y. The average chamber concentration for Experiment 2 was 1960 pp
m. In Experiment 1, the color of the compound in the bubbler grad
ually changed from a clear, colorless liquid to a clear, light gol
d liquid during the exposure. Vapors going into the chamber appea
red to recondense in the chamber intake port and small drops of th
e liquid compound were seen dripping into the port and along one c
orner of the chamber. At 95 minutes into the exposure, the Cellos
eal between the two parts of the trap flask became hot causing the
 top and bottom sections to slide apart. The exposure was stopped
 for nine minutes to replace the t
rap with a new trap flask (1,000 ml three-neck round-bottom flask)
. The exposure was run for an additional nine minutes to compensa
te for the interruption. During the exposure, a total of 906.84 g
 of test substance was delivered in a total volume of 62,280 liter
s of air,yielding a nominal exposure concentration of 14.56 millig
rams per liter or 2,920 ppm. Chamber air concentration measured u
sing the Miran IA Ambient Air Analyzer yielded a mean chamber conc
entration of 3,900 ppm. Subsequent recalibration of the infrared
monitor confirmed the initial calibration. The difference between
 nominal and measured concentration was attributed to differences
between calculated and true chamber air flow rates.

In Experiment 2, vapors going into the chamber appeared to reconde
nse in the chamber intake port and liquid test substance was seen
dripping in small drops into the port and along one corner of the
chamber. During the exposure, a total of 460.63 grams of the test

substance was delivered in a total volume of 62,280 liters of air , yielding a nominal exposure concentration of 7.4 milligrams per liter or 1,480 ppm. Chamber air concentration measured using the Miran IA Ambient Air Analyzer yielded a mean chamber concentration of 1,960 ppm. Subsequent recalibration of the infrared monitor confirmed the initial calibration. The differences between nomina l and measured concentration was attributed to differences between calculated and true chamber air flow rates.

During the exposures, both groups of test animals showed an immedi ate response to the test substance. Experiment 1 showed signs of excessive lacrimation, reduced activity, body tremors, lack of lim b coordination, and insensitivity to sound stimuli. During the 14 -day post-exposure period, 8/10 rats exhibited a lack of support i n limbs at day 3, but all rats recovered by day 4. At day 14, 4/1 0 rats exhibited dry rales while the rest of the rats were normal. Individual body weights for Experiment 1 revealed two female rat s with slower than normal weight gains which may be treatment rela ted. Individual necropsy examinations revealed lung discoloration in 5/10 rats. This finding is not unusual for Sprague-Dawley rat s in this type of exposure.

During Experiment 2, the most frequently noted signs were lack of coordination and lack of limb support. Other signs observed durin g the exposure were squinting, body tremors, and rapid breathing. Upon removal from the chamber and during the two hour post-exposu re observation period the most frequently noted sign was dry rales . Other signs noted at this time were moist rales, yellow stainin g of the anogenital fur, dried material around the nose, and red n asal discharge. These signs were scattered in appearance. The ob servations noted during the exposure appear to indicate an immedia te treatment-related effect of the test substance. The signs, how ever, appeared to be reversible upon removal from the chamber. Du ring the 14-day post-exposure observation period, the most frequen tly noted observation was dry rales. Other signs observed during this period were mucoid nasal discharge and dried red material aro und the nose. All these observations were scattered in appearance and did not reflect treatment-rel
ated effects. At day 14, only 2/10 rats exhibited dry rales; the rest of the animals were normal. Individual body weights appeared normal in all animals. Individual necropsy examinations revealed lung discoloration in 2/10 rats. These findings are not unusual for Sprague-Dawley rats in this type of exposure and do not repres ent any treatment-related effects.","No mortality was observed dur ing two six-hour exposures to vapors of MCTR-51-79 at 3,900 or 1,9 60 ppm (mean, measured concentrations). There were differences be tween nominal and measured concentrations. These differences were attributed to differences between calculated and true chamber air

flow rates.","Acceptable","The key parameters (exposure duration and observations) were appropriate and adequately described in the study.",,"An Acute Inhalation Toxicity Study of MCTR-51-79 in the Rat. Project No.: 79-7281. Bio/dynamics, Inc., East Millstone, New Jersey, January 8, 1980 (M511-79).","Y"
15022002093307.0,1,2/16/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-79-78
Purity: 96%
Additions: None reported
Solvent Carrier: Assume corn oil
Contaminants: None reported
Chemical formula: C9H12",,"Federal Hazardous Substances Act Regula tions (16 CFR 1500)","Yes",1978,"rat","Sprague-Dawley","Both",5,5, "Assume corn oil","Oral","The purpose of this study was to determi ne the acute oral toxicity of the test substance using rats. Fort y healthy albino Sprague-Dawley rats (five males and five females per dose) ranging in body weights between 200 and 300 grams were e mployed. Animals were fasted 18-hours prior to dosing. The test material was administered by oral gavage at dose levels of 3,000, 4,000, 5,000 and 6,000 mg/kg. Animals were observed for mortality and overt signs of toxicity daily for 14 days. Animals that did not survive the observation period were given a necropsy examinati on for gross organ pathology. At the end of the 14 days, survivin g animals were sacrificed and observed for gross organ pathology. Body weights were recorded at study initiation and termination (s urvivors only).","=",4850,"mg/kg-bw","2/10, 3/10, 5/10, and 7/10 d ead for 3,000, 4,000, 5,000 and 6,
000 mg/kg, respectively","Deaths occurred as follows (male and fem ale deaths not separated): 2/10 rats dead at day 3 in 3,000 mg/kg; 2/10 rats dead at day 2 and 1/8 dead at day 5 in 4,000 mg/kg; 1/1 0 rats dead between 6-24 hrs, 2/9 dead at day 2, and 2/7 dead at d ay 3 in 5,000 mg/kg; 1/10 rats dead between 1-3 hrs, 3/9 dead betw een 6-24 hrs, and 3/6 dead at day 2 in 6,000 mg/kg.

Clinical signs of toxicity observed in rats at 3,000 mg/kg include d motor paralysis, motor ataxia and dypsnea (2/10), motor ataxia a nd dypsnea (1/10), and diarrhea (1/10). Onset of signs was observ ed between 6-24 hrs (2/10), day 2 (3/10), and day 3 (1/8). All sur viving rats were normal by day 4 and gained an average of 62 g by the end of the study. Gastritis (1/2), and gastritis and enteriti s (1/2) were observed at necropsy on animals that died during the study. Gastritis (2/8) was observed at autopsy on animals that we re sacrificed at the end of the study. All other rats (6/8) were normal at autopsy. Clinical signs of toxicity observed in rats at 4,000 mg/kg included motor paralysis and dypsnea (2/10), motor at axia, hypoactivity (1/10), and motor paralysis, hyporeactivity, dy psnea, and lethargy (1/10). Onset of signs was observed between 6 -24 hrs (4/10), day 2 (2/8), and day 3 (2/8). All surviving rats

were normal by day 6 and gained an average of 76 g by the end of t
he study. Gastritis and enteritis
(3/3) were observed at necropsy on animals that died during the st
udy. All surviving rats (7/10) were normal at autopsy. Clinical
signs of toxicity observed in rats at 5,000 mg/kg included motor a
taxia, intermittent tonic convulsions, dypsnea, and motor paralysi
s (1/10), motor ataxia, intermittent tonic convulsions, and diarrh
ea (1/10), motor ataxia, motor paralysis, and dypsnea (1/10), moto
r paralysis and dypsnea (2/10), diarrhea (1/10), and motor ataxia
(1/10). Onset of signs was observed between 1-3 hrs (2/10), 3-6 h
rs (1/10), 6-24 hrs (6/9), and day 2 (2/7). All surviving animals
were normal by day 4 and gained an average of 52 g by the end of t
he study. Animals that died during the study exhibited the follow
ing on necropsy: gastritis, enteritis, and intestinal blood vessel
s injected (1/5), gastritis, enteritis, intestinal blood vessels i
njected, and red fluid in bladder (1/5), gastritis, enteritis, int
estinal blood vessels injected, hemmorrhagic stomach, and dark red
fluid in bladder (1/5), and gastr
itis and enteritis (2/5). Animals sacrificed at the end of the st
udy exhibited gastritis and intestinal blood vessels injected (2/5
), gastritis (1/5), or were normal (2/5) on autopsy. Clinical sig
ns of toxicity observed in rats at 6,000 mg/kg included motor para
lysis and dypsnea (4/10), motor paralysis and intermittent tonic c
onvulsions (1/10), motor ataxia, motor paralysis, and dypsnea (1/1
0), and motor ataxia (3/10). Onset on signs was observed between
0-1 hrs (2/10), 1-3 hrs (4/9), 3-6 hrs (4/9), 6-24 hrs (5/6), and
day 2 (1/3). All surviving animals were normal by day 3 and gaine
d an average of 95 g by the end of the study. Animals that died d
uring the study exhibited the following at necropsy: enteritis an
d red fluid in bladder (2/7), gastritis, enteritis, and clear liqu
id in stomach and intestines (1/7), gastritis, enteritis, and clea
r liquid in stomach (1/7), gastritis and enteritis (1/7), gastriti
s, enteritis, and blood vessels of stomach injected (1/7), and ent
eritis (1/7). Surviving animals t
hat were sacrificed at the end of the study exhibited lung discolo
ration (1/3) or were normal (2/3) on autopsy. No apparent sex dif
ferences in mortality and clinical signs were noted.","The acute o
ral LD50 of MCTR-79-78 (Toluene, p-ethyl) in albino Sprague-Dawley
rats was 4,850 mg/kg with 95% confidence limits of 6,062 and 3,88
0 mg/kg. Toluene, p-ethyl (96%) is moderately toxic to Sprague-Da
wley rats. Results indicate that the test substance may cause gas
trointestinal irritation. Based on the clinical signs, the test s
ubstance appears to target the CNS and cause CNS depression.","Acc
eptable","Experimental design and key parameters (number of animal
s/dose, concentrations, number of days observed, etc.) are appropr
iate and adequately described in the study.",,"Evaluation of MCTR-
79-78 1. Acute Oral LD50 Rat. Foster D. Snell, Inc. Project #2
632 Subsidiary of Booz, Allen & Hamilton, Inc., 66 Hanover Road, F

lorham Park, New Jersey 07932, June 8, 1978 (M791-78).","Y"
15022002093307.0,2,2/19/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-79-78
Purity: 96%
Additions: None reported
Solvent Carrier: None
Contaminants: None reported
Chemical formula: C9H12",,"Federal Hazardous Substances Act Regulations (16 CFR 1500.40)","Yes",1978,"rabbit","New Zealand white","Both",5,5,"None","Dermal","The purpose of this study was to evaluate the acute dermal toxicity of the test substance using New Zealand White rabbits. Ten healthy New Zealand White rabbits (2.3 to 3.0 kg; 5 males and 5 females) were dosed dermally with a single application of the test substance at a dose of 5,000 mg/kg. The trunk of each animal was clipped free of hair prior to application of the test substance. Four of the rabbits, 2 males and 2 females, were further prepared by abrading the test site. Epidermal incisions every two or three centimeters were made longitudinally over the area of exposure. The incisions were sufficiently deep to penetrate the stratum corneum, but not to disturb the derma or elicit bleeding. The test substance was held in contact with the skin for 24 hrs by means of a non-reactive,heavy guage plastic covered with an opaque wrapping. At the end
of the 24-hr exposure period, the wrappings were removed and the skin was gently wiped to remove any remaining test substance. Animals were observed for mortality and overt signs of toxicity during the day of dosing and at least once daily for 14 days. Animals not surviving the observation period were given a necropsy examination for gross organ pathology. At the end of the 14-day observation period, surviving animals were sacrificed and observed grossly for organ pathology. Body weight data was recorded initially and, for survivors, at termination of the study.",">",5000,"mg/kg-bw","No mortalities were observed in the study.","All animals survived to the end of the study. All animals exhibited moderate erythema upon removal of wrappings. Animals sacrificed at the end of the 14-day observation period exhibited the following: Test skin site - several small ulcerations (6/10), several small ulcerations and moderate erythema (1/10), normal (3/10); Internal - subdermal blood vessels injected and lungs discol
ored (1/10), blood vessels of stomach injected (1/10), subdermal blood vessels injected (2/10), subdermal blood vessels injected and both kidneys discolored (1/10), left kidney partially discolored and hardened (interior of hardened tissue granular) (1/10), normal (4.10). Body weight gain over the 14-day period averaged 0.02 kg. Females on average gained 0.14 kg while males lost an average of 0.1 kg.","In accordance with the Federal Hazardous Substances Act Regulations 16 CFR 1500.3, the test substance, MCTR-79-78, was not toxic by the dermal route. The acute dermal LD50 to New Zealand

White rabbits was >5,000 mg/kg.","Acceptable","Experimental desig
n and key parameters (number of animals, number of days observed,
etc.) are apppropriate and adequately described in the study.",,"E
valuation of MCTR-79-78 2. Acute Dermal Toxicity, Rabbit. Foste
r D. Snell, Inc. Project #2632. Subsidiary of Booz, Allen & Hami
lton Inc., 66 Hanover Road, Florham Park, New Jersey 07932, June
8, 1978 (M792-78).","Y"
15022002093307.0,3,2/19/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-79-78
Purity: 96%
Additions: None reported
Solvent Carrier: None
Contaminants: None reported
Chemical formula: C9H12",,"Federal Hazardous Substances Act Regula
tions (16 CFR 1500.41)","Yes",1978,"rabbit","New Zealand white","M
",6,0,"None","Dermal","The purpose of this study was to evaluate t
he primary dermal irritation of the test substance using New Zeala
nd White rabbits. The dorsal trunks of six healthy New Zealand Wh
ite rabbits (sex and age were not reported) were clipped free of h
air. One side of each animal was further prepared by abrading the
skin. Four incisions were made in a cross-hatch to serve as the
abraded test site. The incisions broke the stratum corneum but di
d not disturb the derma or elicit bleeding. Each animal recieved
two 0.5 ml applications of the test substance, one on the intact s
kin site and the other on the abraded skin site (0.5 g/site; speci
fic gravity 1 g/ml). Surgical gauze (2 inch x 2 inch) was applied
to the treatment sites and secured with adhesive tape. The entir
e trunk was then encased in a heavy gauge plastic cuff. The test
substance remained in contact with
the skin for 24 hrs after which the plastic cuff and gauze were re
moved. Treated skin sites were scored for irritation (see below)
24 and 72 hrs after application of the test substance.

The scale for rating skin reactions is as follows:
Erythema and Eschar Formation
0=no erythema
1=very slight erythema (barely perceptible)
2=well defined erythema
3=moderate to severe erythema
4=severe erythema (beet redness) to slight eschar formation (injur
ies in depth)

Edema Formation
0=no edema
1=very slight edema (barely perceptible)
2=slight edema (edges of area well defined by definite raising)
3=moderate edema (raised approximately 1 mm)
4=severe edema (raised more than 1 mm and extending beyond area of

exposure)

Draize, H.J., in ""Appraisal of the Safety of Chemicals in Foods, Drugs, and Cosmetics"", Assoc. Food and Drug Officials of the U.S. , Austin, Texas, 1959.",">",500,"mg/site","No deaths were observed .","No deaths or clinical signs were observed. At 24 hrs, the ave rage erythema score for both intact and abraded animals was 1.50, and the average edema score for both intact and abraded animals wa s 0.67. The combined erythema and edema average score at 24 hrs f or both intact skin and abraded skin was 2.17. At 72 hrs, the ave rage erythema score for intact and abraded animals was 1.50 and 1. 83, respectively, and the average edema score for intact and abrad ed animals was 0.17 and 0.67, respectively. The combined average erythema and edema score at 72 hrs for intact skin and abraded ski n was 1.67 and 2.50, respectively. The primary dermal irritation index was 2.13 (8.51/4).","The Primary Dermal Irritation Index for MCTR-79-78 was 2.13. The undiluted product caused well-defined i nflammation (erythema skin reactio
n values of 2) during the study period. The test substance is cla ssified as moderately irritating as described in 16 CFR 1500.3.","Acceptable","The key parameters (number of animals used, methodolo gy) was appropriate and adequately described in the study.",,"Eval uation of MCTR-79-78 4. Primary Dermal Irritation Rabbit. Foster D. Snell, Inc. Project #2632 Subsidiary of Booz, Allen & Hamilto n, Inc., 66 Hanover Road, Florham Park, New Jersey 07932, June 8, 1978 (M794-78).","Y"
15022002093307.0,4,2/20/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-79-78
Purity: 96%
Additions: None reported
Solvent Carrier: None
Contaminants: None reported
Chemical formula: C9H12",,"Federal Hazardous Substances Act Regula tions (16 CFR 1500.42)","Yes",1978,"rabbit","New Zealand white","M ",6,0,"None","Eye","The purpose of this study was to evaluate the ocular irritation of the test substance using New Zealand White ra bbits. Six healthy New Zealand White rabbits (sex and age were no t reported) without ocular defects were used. Each animal receive d 0.1 ml (0.1 g/eye; specific gravity 1 g/ml) of the test substanc e in one eye. Eyes were observed for the presence of injury to th e cornea, iris, and conjunctivae. Observations were conducted at 1, 24, 48, 72, 96, and 168 hrs after instillation of the test subs tance.

The injuries were assigned a numerical score according to the ""I llustrated Guide for Grading Eye Irritation Caused by Hazardous Su bstances"", U.S. Consumer Product Safety Commission, Washington, D .C., as presented below.

Cornea
0=no ulceration opacity
(1)*=scattered or diffuse areas of opacity (other than slight dull
ing of normal luster), details of iris clearly visible
2=easily discernible translucent areas, details of iris slightly o
bscured
3=nacreous areas, no details of iris visible, size of pupil barely
discernible
4=complete corneal opacity, iris not discernible

Iris
0=normal
(1)*=markedly deepened folds, congestion, swelling, moderate circu
mcorneal injection (any of these or combination of any thereof), i
ris still reacting to light (sluggish reaction is positive)
2=no reaction to light, hemorrhage, gross destruction (any or all
of these)

Conjunctivae
(A) redness (referes to palpebral and bulbar conjunctivae excludin
g cornea and iris)
0=vessels normal
1=some vessels definitely injected
(2)*=diffuse, crimson red, individual vessels not easily discernib
le
3=diffuse beefy red

(B) Chemosis
0=no swelling
1=any swelling above normal (includes nictitating membrane)
(2)*=obvious swelling with partial eversion of lids
3=swelling with lids about half closed
4=swelling with lids more than half closed

(C) Discharge
0=no discharge
1=any amount different from normal (does not include small amounts
observed in inner canthus of normal animals)
2=discharge with moistening of the lids and hairs just adjacent to
the lids
3=discharge with moistening of lids and hairs, and considerable ar
ea around the eye

*Bracketed figures indicate lowest grades considered positive unde
r the Federal Hazardous Substances Act Regulations at 16 CFR 1500.
42.

An animal shall be considered as exhibiting a positive reaction if the test substance produces at any of the readings ulceration of the cornea (other than fine stippling), or opacity of the cornea (other than a slight dulling of the normal luster), or inflammation of the iris (other than slight deepening of the folds, or rugae, or a slight circumcorneal injection of the blood vessels), or if such substance produces in the conjunctivae (excluding the cornea and iris) an obvious swelling with partial eversion of the lids or a diffuse crimson red with individual vessels not easily discerni ble. The test shall be considered positive if four or more of the animals in the test group exhibit a positive reaction. If only o ne animal exhibits a positive reaction, the test shall be regarded as negative. If two or three animals exhibit a positive reaction , the test is repeated using a different group of six animals. Th e second test shall be considered positive if three or more of the animals exhibit a positive reacti
on. If only one or two animals in the second test exhibit a posit ive reaction, the test shall be repeated with a different group of six animals. Should a third test be needed, the substance will b e regarded as an irritant if any animal exhibits a positive reacti on.",">",100,"mg/eye","No deaths were observed.","The cornea and t he iris were normal in all animals throughout the observation peri od. The grades of ocular reaction for redness of the conjunctivae for the six animals were as follows:
1 hr - 1,1,2,1,1,2; 24 hrs - 2,2,3,3,2,3; 48 hrs - 2,2,3,2,2,3; 72 hrs - 2,1,2,2,1,3; 96 hrs - 2,0,2,2,1,3; 168 hrs - 1,0,2,2,0,3. The grades of ocular reaction for chemosis of the conjunctivae wer e as follows: 1 hr - 0,0,0,0,0,0; 24 hrs - 1,0,2,1,1,2; 48 hrs - 1 ,0,1,1,1,2; 72 hrs - 1,0,1,1,1,1; 96 hrs - 0,0,1,1,0,1; 168 hrs - 0,0,1,0,0,1. The grades of ocular reaction for discharge of the c onjunctivae were as follows: 1 hr - 0,0,0,0,0,0; 24 hrs - 1,0,1,0, 0,1; 48 hrs - 1,1,1,0,0,1; 72 hrs - 1,0,0,0,0,1; 96 hrs - 0,0,0,0, 0,1; 168 hrs - 0,0,0,0,0,0.

The average eye irritation scores are as follows: 1 hr = 5; 24 hrs = 12; 48 hrs = 12; 72 hrs = 9; 96 hrs = 7; 168 hrs = 6. These sco res were obtained as follows: the conjunctivae score is 4 times th e sum of the grades for redness plus 2 times the sum of the grades for chemosis. This number is divided by 6 to obtain the average eye irritiation scores for each time period.","In accordance with the Federal Hazardous Substances Act Regulations (16 CFR 1500.3), the test substance, MCTR-79-78 is classified as slightly irritatin g (Category 2A eye irritant). The test substance caused moderate c hemical conjunctivitis. Redness of the conjunctivae decreased in severity but was still evident at day 7. The cornea and iris were normal throughout the observation period.","Acceptable","The key parameters (number of animals used, methodology) was appropriate a nd adequately described in the study.",,"Evaluation of MCTR-79-78

3. Ocular Irritation Rabbit. Foster D. Snell, Inc. Project #263
2 Subsidiary of Booz, Allen & Ham
ilton, Inc., 66 Hanover Road, Florham Park, New Jersey 07932, Jun
e 8, 1978 (M793-78).","Y"

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","GLP","Year","Species","Strain","Sex","NumberofMales","NumberofFemales","Route","ExposPeriod","Frequency","Doses","ControlGroup","StatMeth","MethodRem","MatNPrec","MatNOEL","MatNUnit","MatNEffect","MatLPrec","MatLOEL","MatLUnit","MatLEffect","DevNPrec","DevNOEL","DevNUnit","DevNEffect","DevLPrec","DevLOEL","DevLUnit","DevLEffect","ActualDose","MaternalData","FetalData","StatResults","ResultsRem","ConcludingRem","Reliability","ReliRem","GeneralRem","RefRem","Completed"
15022002093307.0,1,3/13/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: Sample-01038003 (MCTR-304-79)
Purity: Assume 100% for dose calculations (actual 96%)
Additions: None reported
Solvent Carrier: Mazola corn oil (5 ml/kg)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.3700","Yes",1981,"rat","CD-1","F",0,25,"Oral","6-19","daily","0, 25, 100, and 200 mg/kg/day","Yes","Bartlett's test, Chi-square, ANOVA, Dunnett's multiple comparison, Mann-Whitney U-test (p<0.05 and p<0.01)","The purpose of this study was to determine the teratogenic potential of the test substance to rats. One hundred untreated, sexually mature, virgin female Charles River COBS CD rats were used. These rats were approximately 12 weeks old at the time of mating and had been acclimated to laboratory conditions for 13 days prior to study initiation.

One female and one male rat of the same strain and source were placed together for mating. The occurrence of copulation was determined by daily inspection for a copulatory plug. The day that evidence of mating was detected was designated day 0 of gestation and the female was returned to an individual cage.

Mated females were consecutively assigned in a block design to one control and three treatment groups consisting of 25 rats each. The appropriate amount of Sample-01038003 was added to 50 ml of the vehicle, Mazola corn oil, and mixed by hand to ensure a homogeneous mixture. The test substance was prepared fresh daily and administered by oral gavage as a single daily dose to pregnant rats on days 6 through 19 of gestation. Dosage levels were 0 (corn oil control, 5 ml/kg), 25, 100, and 200 mg/kg/day (at a constant volume of 5 ml/kg).

Prior to treatment, the dams were observed daily for mortality and overt changes in appearance and behavior. They were observed daily for mortality and clinical signs of toxicity on days 6 through 20 of gestation. Individual maternal body weights were recorded on gestation days 0, 6, 9, 12, 16, and 20. A Cesarean section was performed on each female on gestation day 20 immediately following sacrifice by carbon dioxide inhalation. The uterus was excised and weighed prior to removal of the fetuses. The number and location of viable and nonviable fetuses, early and late resorptions, total implantations and corpora lutea were recorded. The thoracic and abdominal cavities and organs of the dams were examined for grossly evident morphological changes and the carcasses discarded. Uteri from females that appeared nongravid were opened and placed in 10% ammonium sulfide solution for confirmation of pregnancy status.

All fetuses were individually weighed and examined for external malformations and variations, including the palate and eyes. Each fetus was externally sexed and individually numbered and tagged for identification. Approximately one-half of the fetuses were placed in Bouin's fixative for subsequent visceral examination by razor blade sectioning. The remaining one-half of the fetuses were fixed in alcohol, macerated in potassium hydroxide, and stained with Alizarin Red S for subsequent skeletal examination.

All statistical analyses compared the treatment groups to the control group with the level of significance at p<0.01 and p<0.05. The male to female fetal sex distribution and the number of litters with malformations were compared using the Chi-square test criterion with Yates' correction for 2 x 2 contingency tables and/or Fisher's exact probability tests to judge significance of differences. The number of early resorptions and postimplantation losses were compared by the Mann-Whitney U-test. The mean number of viable fetuses, total implantations, corpora lutea, and mean fetal body weights were compared by ANOVA, Bartlett's test for homogeneity of variances and Dunnett's multiple comparison tables to judge significance of differences.",">",200,"mg/kg-bw","No statistically significant effects were observed",">",200,"mg/kg-bw","No statistically significant effects were observed",">",200,"mg/kg-bw","No statistically significant effects were observed",">",200,"mg/kg-bw","No statistically significant effects were observed","All doses were based on nominal concentrations.","No statistically significant effects were observed at any dose level.","No statistically significant effects were observed at any dose level.","No statistically significant effects were observed at any dose level.","There were no biologically meaningful differences in the appearance or behavior of the rats in the 25, 100, or 200 mg/kg/day groups when compared to controls. Hair loss (primarily of the limbs and abdomen) occurred with similar frequency in all treatment and control groups at various intervals throughout the treatment period. Survival was 100% in the control and all Sample-01038003 treated groups.

Hydrometra and hydronephrosis occurred infrequently in the control and treatment groups. An abscess of the spleen and pancreatitis were observed in one control animal and white caseous material in the pericardial sac was observed in another control animal. Peric arditis was found in one animal in the 100 mg/kg/day group and pit ted kidneys were found in one animal in 200 mg/kg/day. No other p ostmortem abnormalities were observed in any of the study animals.

There were no biologically meaningful differences in mean maternal body weight gain throughout the entire gestation period in any tr eated group when compared to controls. The mean maternal adjusted body weight gain (dam body weight exclusive of the uterus and con tents) in all treatment groups was also comparable to the control group during this interval. In the pilot teratology study (see Re ference), a moderate reduction in maternal body weight gain was no ted at the 100 mg/kg/day group. This apparent difference in toxic ity may be attributable, in part, to the use of corn oil as a vehi cle in the present study, whereas the test substance was administe red without vehicle in the pilot study.

There were no biologically meaningful or statistically significant differences in the mean numbers of corpora lutea, total implantat ions, early resorptions, postimplantation loss, viable fetuses, t he fetal sex distribution or mean fetal body weight in any of the treated groups when compared to the control group. Nonviables and late resorptions were not observed in the control or in any of th e treated groups. A slight increase in mean postimplantation loss was observed in the 25 mg/kg/day group when compared to controls. However, no dose-related trend was evident and this response was considered to be due to a random occurrence. Mean postimplantati on loss number for 0, 25, 100, and 200 mg/kg/day was 13, 29, 19, a nd 19, respectively.

There were no biologically meaningful or statistically significant differences in the number of litters with malformations in any of the treated groups compared to controls. Microphthalmia and thor acoschisis each occurred in one fetus in one litter in the 25 and 200 mg/kg/day groups, respectively. Scoliosis was observed in one litter from both of the 200 mg/kg/day and control groups. The nu mber of litters (and fetuses) with genetic and developmental varia tions in the treated groups was comparable to controls.","Pregnant Charles River COBS CD rats were used to determine the teratogenic potential of Sample-01038003. Dosage levels of 25, 100, and 200 mg/kg/day were administered orally by gavage as a single daily dos e on days 6 through 19 of gestation at a constant volume of 5 ml/k g. The control group received the vehicle only, Mazola corn oil, on a comparable regimen. Cesarean sections were performed on all females on gestation day 20.

Survival was 100% in all dosage groups. There were no biologicall y meaningful differences in appearance, behavior or mean maternal body weight gain of rats in any of the treated groups when compare d to controls. There were no biologically meaningful or statistic ally significant differences in the mean numbers of corpora lutea, total implantations, early resorptions, postimplantation loss, vi able fetuses, the fetal sex distribution, mean fetal body weight o r the number of litters with malformations in any of the treated g roups when compared to controls. The number of litters (and fetus es) with genetic and development variations in the treated groups was also comparable to controls.

Treatment with Sample-01038003 did not produce a teratogenic respo nse when administered orally, in a corn oil vehicle, to pregnant r ats at a dosage of 200 mg/kg/day or less.","Acceptable","All key p arameters (i.e., number of animals, observations, doses, etc.) wer e appropriate and adequately described in the study.",,"Teratology Study in Rats (MCTR-304-79). International Research and Developme nt Corporation, Mattawan, Michigan, October 15, 1981 (M3040-79).

A pilot teratology study was conducted to establish dosage levels for the present study (Pilot Teratology Study in Rats, MCTR-310-79 , International Research and Development Corporation, December 11, 1980 (M3100-79). The study was evaluated and the synopsis is as follows: Pregnant Charles River COBS CD rats were used to determin e dosage levels of Sample-01038003 for a teratology study (M3040-7 9). Dosage levels of 100, 300, 750, 1500, and 3000 mg/kg/day were administered orally by gavage as a single daily dose on days 6 th rough 19 of gestation, at volumes of 0.116, 0.349, 0.872, 1.744, a nd 3.488 ml/kg, respectively. The control group received distille d water only on a comparable regimen at a volume of 3.488 ml/kg. Uterine examinations were performed on all surviving dams on gesta tion day 20. There were no biologically meaningful differences in appearance or behavior or mean uterine examination values in the 100 mg/kg/day treatment group when compared to controls. All rats in the 3000 mg/kg/day group, four
rats in the 1500 mg/kg/day group, and one rat in the 750 mg/kg/day group died prior to the scheduled sacrifice date. The cause of d eath for all of these rats could not be determined at necropsy exa mination.

Postmortem findings in the 1500 and 3000 mg/kg/day groups included inflammation and reddening of the gastrointestinal mucosa and ero sions of the stomach lining. Antemortem findings included stained or matted haircoat and dried red or brown matter around the nose and mouth. A severe decrease in mean maternal body weight gain oc curred in the 300, 750, and 1500 mg/kg/day groups and a moderate r eduction in mean maternal body weight gain was noted in the 100 mg

/kg/day treatment group over the entire treatment period. Uterine examination findings revealed an increase in the mean number of e arly resorptions in the 300, 750, and 1500 mg/kg/day treatment gro ups with a corresponding increase in mean postimplantation loss in these treatment groups when compared to controls.

Based on these results, a dosage level of 300 mg/kg/day would be c onsidered excessive for a teratology study in rats with Sample-010 38003.

This study is acceptable based the purpose and the key parameters (i.e., dosages, number of animals, observations, etc.).",
15022002093307.0,2,3/14/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: PET; Sample-01038003 (MCTR-305-79)
Purity: Assume 100% for dose calculations (actual 97%)
Additions: None reported
Solvent Carrier: Mazola corn oil (0.5 ml/kg)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.3700","Yes",1981," rabbit","Dutch Belted","F",0,16,"Oral","6-27","Daily","0, 25, 125, 200, and 250 mg/kg/day","Yes","Bartlett's test, Chi-square, ANOVA , Dunnett's multiple comparison, Mann-Whitney U-test (p<0.05 and p <0.01)","The purpose of this study was to determine the teratogeni c potential of the test substance to rabbits. Sexually mature vir gin female Dutch Belted rabbits (5-8 months old) were placed on th e study following a detailed observation and a 30-day acclimation period. During the initial phase of the acclimation, stool sample s from each rabbit were examined for ova or parasites. The rabbit s (16 per treatment) were randomly assigned by a computer-generate d program to three treatment groups (25, 125, and 250 mg/kg/day) a nd vehicle control. Because of unanticipated maternal toxicity at 250 mg/kg/day (8/16 dead by day 15), an additional dose, 200 mg/k g/day, was added along with an additional control group (Control I I). These females were artificial
ly inseminated with diluted semen from proven males of the same st rain and source. Ovulation was induced by an injection of human c horionic gonadotropin (HCG, 100 U.S.P. Units) into the marginal ea r vein within one hour following insemination. The day of insemin ation was designated day ""0"" of gestation.

The test substance was administered orally by gavage as a single d aily dose. Test substance administration began on day 6 and conti nued up to and including day 27 of gestation. Individual dosages were based on gestation day 6 body weights. Dosing solutions of t he test substance were freshly prepared in corn oil daily. Prior to test substance administration, the rabbits were observed daily for mortality and overt changes in appearance and behavior. The f emales were observed daily for mortality and clinical signs of tox icity from gestation days 6 through 28. The dams were weighed on gestation days 0, 6, 12, 18, 24, and 28.

On the 28th day of gestation, all surviving females were sacrifice d by an overdose of sodium pentobarbital via the marginal ear vein . Immediately following sacrifice, the uterus was excised and wei ghed prior to removal of the fetuses. The number and location of viable and nonviable fetuses, early and late resorptions, and the number of total implantations and corpora lutea were recorded. Th e abdominal and thoracic cavities and organs of the dams were exam ined for grossly evident morphological changes and the carcasses d iscarded. Uteri from females that appeared nongravid were opened and placed in 10% ammonium sulfide solution for confirmation of pr egnancy status.

All fetuses were individually weighed and examined for external ma lformations and variations, including the palate and eyes. Each f etus was dissected, internally sexed, and examined for visceral ma lformations and variations, including the brain by a mid-coronal s lice. The eviscerated, skinned fetuses were individually numbered and tagged for identification, fixed in alcohol, macerated in pot assium hydroxide, and stained with Alizarin Red S for subsequent s keletal examination. Fetal findings were classified as malformati ons or genetic or developmental variations.

All statistical analyses compared the treatment groups to the cont rol group, with the level of significance at p<0.05 and p<0.01. T he male to female fetal sex distribution and the number of litters with malformations were compared using the Chi-square test criter ion with Yates' correction for 2 x 2 contingency tables and/or Fis her's exact probability test to judge significance of differences. The number of early and late resorptions and postimplantation lo ss were compared by the Mann-Whitney U-test to judge significance of differences. The mean number of viable fetuses, total implanta tions, corpora lutea and mean fetal body weights were compared by ANOVA, Bartlett's test for homogeneity of variances and Dunnett's multiple comparison tables to judge significance of differences.", "=",200,"mg/kg-bw","mortality","=",250,"mg/kg-bw","mortality","=", 200,"mg/kg-bw","No treatment-related effects observed.",">",200,"m g/kg-bw","Mortality at 250 mg/kg/day prevented comparisons","All d oses were based on nominal concent
rations.","The number of dead aminals in control I, 25, 125, and 2 50 mg/kg/day were 2, 1, 3, and 12, respectively.","No effect relat ed to treatment on Cesarean section parameters or the number of fe tuses (litters) with malformations were observed at 25, 125, or 20 0 mg/kg/day.","Significant differences in maternal body weights we re observed in control I and 125 mg/kg/day groups (p<0.05). These differences were not considered biologically meaningful.","All mo

rtality in this study occurred during the treatment period. Morta lity (dead/total) for control (I), control (II), 25, 125, 200, and 250 mg/kg/day were 2/16, 0/16, 1/16, 2/16, 0/16, and 11/16, respe ctively. The number aborted and sacrificed were 1, 0, 1, 3, 2, an d 1, respectively. The number aborted and died were 0, 0, 0, 1, 0 , and 1, respectively. The number surviving to sacrifice at study termination were 13, 16, 14, 10, 14, and 3, respectively. The ca use of death for two females in the 250 mg/kg/day group was determ ined to be due to an intubation er
ror. Prior to death, a reduced amount of feces beneath the cage w as noted in a majority of animals in the treatment and control gro ups that died. Additional observations prior to death were labore d breathing, moribund behavior, limp and lethargic appearance, and emaciation. Upon necropsy examination, focal erosion of the stom ach mucosa and discolored mucoid intestinal contents were observed in these animals. The uterine contents of these dams were primar ily late resorptions in the control (I), 25, and 125 mg/kg/day gro ups, and normally developing implantations in the 250 mg/kg/day gr oup. One dam in the 125 mg/kg/day group apparently aborted at som e time during gestation as two empty implantation sites were obser ved upon necropsy examination; uterine contents also included one early resorption and one normally developing implantation.

During the treatment period, reduced amounts of feces beneath the cage were noted in a majority of the females on the study and occu rred with similar frequency among the treated and control groups (I and II). Hair loss (primarily of the forelimbs and ventral regi on), nasal and ocular discharges, and matting of the haircoat (pri marily in the nasal and ocular regions) were also observed with si milar frequency among the treatment and both control groups.

Upon necropsy examination at Cesarean section, lung congestion, hy droceles on the oviduct(s) and pitted kidneys were observed in a f ew animals in the treated and control groups. These findings were considered as spontaneously-occurring in rabbits of this age and strain and not a consequence of treatment. A solid circumscribed area within the azygous lobe of the lung (diagnosed as multifocal abscesses at microscopic examination), lungs one-third normal size , and a heart one-half normal size were observed in one female in the 250 mg/kg/day group.

Mean maternal body weight losses in the control (I) and 125 mg/kg/ day groups and no gain in the 25 mg/kg/day group occurred during t he treatment period (gestation days 6 to 28). The reason for thes e losses is unclear. However, given the variability of maternal b ody weight gain in this species, the effect was probably not a con sequence of treatment as control groups also displayed losses and no apparent dose-response relationship was noted. Adjusted matern

al body weight change (gestation day 0 to 29; with gestation day 2 9 body weight minus gravid uterus and contents) showed comparable losses in the control (I), 25 and 125 mg/kg/day groups. In view o f the excessive mortality which occurred at the 250 mg/kg/day grou p, comparisons of mean maternal body weight gain during treatment and gestation could not be made; body weight loss was noted prior to death in a majority of animals at this dose level. At the 200 mg/kg/day level, mean maternal body weight gain exceeded control g roup (II). Mean maternal body wei
ght change (0 to 28 days of gestation; grams) for control (I), con trol (II), 25, 125, 200, and 250 mg/kg/day was 13, 219, 34, 22, 34 3, and -9, respectively. Adjusted weight change was -231, 4, -240 , -263, 50, and -198, respectively.

There were no biologically meaningful or statistically significant differences in the mean numbers of corpora lutea, total implantat ions, early or late resorptions, postimplantation loss, viable fet uses, the fetal sex ratio or mean fetal body weight in the 25 or 1 25 mg/kg/day groups when compared to control (I). A slight increa se in mean postimplantation loss occurred at the 25 mg/kg/day leve l. However, since this value (1.1) fell within the range of the h istorical control (0.2 - 1.5) and since the finding was not eviden t at 125 or 200 mg/kg/day levels, the effect was not considered tr eatment-related. Similarly, a decrease in mean fetal body weight was observed in the 125 mg/kg/day group. This value was within th e range of the historical control and may have been due to the sli ght increase in the number of viable fetuses/dam at this level whe n compared to control (I). Meaningful comparisons of Cesarean sec tion parameters could not be made in the 250 mg/kg/day group due t o the severely reduced sample size
as a result of mortality. The mean number of corpora lutea, total implantations, early resorptions, postimplantation loss, viable f etuses, and the fetal sex ratio for the 200 mg/kg/day group were s tatistically comparable to the control group (II). A significant ($p<0.05$) reduction in mean fetal body weight occurred in the 200 m g/kg/day group when compared to the control group (II). However, this value was within the range of historical control and may have been due to the increase in the number of viable fetus/dam seen a t the 200 mg/kg/day group. Mean fetal body weights (grams) for co ntrol (I), control (II), 25, 125, 200, and 250 mg/kg/day were 32.3 , 35.4, 32.3, 28.5, 30.7 (significant from control (II) at $p<0.05$) , and 26.2 (significance not determined due to low sample size).

There were no biologically meaningful or statistically significant differences in the number of litters with malformations in the 25 or 125 mg/kg/day group when compared to the control group (I) or in the 200 mg/kg/day group when compared to the control group (II) . Two malformations (coloboma and atlas-occipital defect) occurre

d as single incidences (on a per fetus basis, only one litter was affected) in the 200 mg/kg/day group; these malformations have not been cited in the historical control. However, their low frequen cy of occurrence in this study negates a possible compound-related effect. The majority of the remaining malformations observed in this study occurred either as single incidents or without an appar ent dose-related trend. Therefore, they were deemed to be spontan eously occurring and not a result of treatment. The number of fet uses (and litters) with developmental and genetic variations in th e 25 and 125 mg/kg/day groups were comparable to control group (I) and historical data. The increas
e in the number of fetuses with one variation (13th full ribs) occ urred in the 125 mg/kg/day group which slightly exceeded the range of historical control values. However, since the number of fetus es affected at the 200 mg/kg/day level was comparable to historica l values, this finding was considered unrelated to treatment. An increase in the number of fetuses with 13th rudimentary ribs (whic h slightly exceeded the range of historical control values) was no ted in the 200 mg/kg/day group; however, this was classified as a skeletal variant, not a malformation. Skeletal examination of int act fetuses from dams in the control group (which aborted on gesta tion day 28) revealed no malformations or genetic or developmental variations. At the 250 mg/kg/day level, meaningful comparisons o f fetal malformations and variations data could not be made due to the reduced number of fetuses available for evaluation.","Eightee n rabbits died during treatment. Mortality in control (I), 25, 12 5, and 250 mg/kg/day were 2, 1, 3,
and 12, respectively. Survival in the 200 mg/kg/day and control g roup (II) was 100%. The cause of death was determined to be an in tubation error for two animals in the 250 mg/kg/day group but coul d not be determined for the remaining animals. Mean maternal body weight losses in the control (I) and 125 mg/kg/day groups and no gain in the 25 mg/kg/day group occurred during the treatment perio d. The reason for these losses is unclear. However, given the so mewhat erratic nature of maternal body weight gain in this species , the effect was probably not a consequence of treatment as contro l group (I) also displayed losses and mean maternal body weight at 200 mg/kg/day level exceeded the control group (II). Females wer e physiologically stressed at 125 and 200 mg/kg/day based on the n umber of spontaneous abortions; 1 and 4 for control (I) and 125 mg /kg/day, respectively, and 0 and 2 for control (II) and 200 mg/kg/ day, respectively. The genetic differences of the rabbits apparen tly contributed to the variation w
ithin and between these groups. Under these particular testing co nditions where the daily dose is administered all at once via oral gavage, even relatively small quantities of a material are capabl e of disturbing the delicate maternal-fetal balance (Khera, Fundam . Appl. Toxicol., 1, 13, 1981). No effect related to treatment on

Cesarean section parameters or the number of fetuses (litters) with malformations occurred in the 25 or 125 mg/kg/day group when compared to control group (I) or in the 200 mg/kg/day when compared to control group (II). There was an increase in the occurrence of one genetic and developmental variation (13th rudimentary ribs) in the 200 mg/kg/day group when compared to the control (II) and historical control values. However, this was considered a skeletal variant and not a malformation. Maternal stress and embryo-toxicity attributable to maternal treatment at high doses have been associated with reports of extra ribs (Kimmel and Wilson, Teratology, 8, 309, 1973). However, when extr
a ribs represent the only positive finding in a teratology study under these conditions, the biological significance is usually low (Khera, Fd. Cosmet. Toxicol., 12, 471, 1974; Hudak and Unguary, Toxicology, 11, 55, 1978). At the 250 mg/kg/day level, meaningful comparisons of these parameters could not be made due to excessive mortality and the subsequent severe reduction of the sample size.

Paraethyl Toluene (PET) did not produce a teratogenic response when administered orally to pregnant rabbits at a dose level of 200 mg/kg/day or less.","Acceptable","All key parameters (i.e., doses, number of animals, observations, etc.) were appropriate and adequately described in the study.",,"Teratology study in Rabbits (MCTR-305-79). International Research and Development Corporation, November 16, 1981 (M305-79).

Pilot Teratology Study in Rabbits (3120-79), reported 12-14-81 by International Research and Development Corporation: Twenty pregnant Dutch Belted rabbits, randomly assigned to one control and three treatment groups of five rabbits each were used in this pilot study to determine dosage levels of PET for a teratology study. Dose levels of 25, 50, and 100 mg/kg/day prepared in corn oil were administered orally by gavage as a single daily dose on day 6 through 27 of gestation at a constant volume of 0.5 ml/kg. The control group received 0.5 ml/kg of the corn oil vehicle on a comparable regimen. Uterine examinations were performed on all surviving animals on gestation day 28. Survival was 100% in the 25 and 50 mg/kg/day groups. Five rabbits died during the treatment period; two in the control and three in the 100 mg/kg/day groups; one dam aborted prior to death at the 100 mg/kg/day level. Upon necropsy, an intubation error was determined as the cause of death for one of the 100 mg/kg/day group females; the
cause of death for the remaining rabbits could not be determined. There were no biologically signficant differences in mean maternal body-weight gain or mean uterine examination observations in the 25 or 50 mg/kg/day groups when compared to the control group. A decrease in the number of total implantations and an increase in the number of postimplantation losses, with a corresponding decreas

e in the number of viable fetuses, was observed at the 100 mg/kg/d ay level. These values were slightly outside the range of the his torical control because one female (of the two examined) had only two implantations; the finding, therefore, is not considered to be biologically significant. The remaining parameters evaluated at uterine examination in this group and mean maternal body-weight ga in were comparable to the control group. Because it appeared that embryotoxicity was not achieved at these dose levels and the evid ence for maternal toxicity was equivocal (due to the death of two controls animals), doses for the d
efinitive study were selected above the range of those used in thi s preliminary study.

Teratology Study in Rabbits on p-Ethyltoluene (1501-80), reported 12-23-81 by International Research and Development Corporation: Pr egnant Dutch Belted rabbits were used in this pilot study to dete rmine dosage levels of PET for a teratology study. Dosage levels of 0, 20, 40, 60, 100, and 200 mg/kg/day prepared in corn oil were administered orally by gavage as single daily doses on days 6 thr ough 27 of gestation at a constant volume of 0.5 ml/kg, to groups of 5 dams each. The control group received 0.5 ml/kg of the corn oil vehicle, on a comparable regimen. Two additional groups of pr egnant rabbits were similarly dosed; one each at 400 and 800 mg/kg /day, at a constant volume of 1 ml/kg. Uterine examinations were performed on all surviving females on gestation day 28. There wer e no biologically significant differences observed in the appearan ce or behavior, mean maternal body-weight gain or mean uterine exa mination observations of rabbits in 20, 40, 60, 100, or 200 mg/kg/ day groups when compared to the hi
storical control group. Survival in these groups was 100%. All r abbits at the 400 and 800 mg/kg/day level died during the treatmen t period: the cause of death was not determined at necropsy for an y of these rabbits. All dams in the 400 mg/kg/day group that surv ived to the first post-treatment weighing showed marked body-weigh t losses. Based on these results, a dosage level of 400 mg/kg/day would be considered excessive for a teratology study.",

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","GLP",
"Year","Species","Strain","Sex","NumberofMales","NumberofFemales",
"Route","ExposPeriod","Frequency","Doses","ControlGroup","PostObsP
eriod","StatMeth","MethodRem","NPrec","NOAEL","NUnit","NEffect","L
Prec","LOAEL","LUnit","LEffect","ActualDose","ToxicResp","StatResu
lts","ResultsRem","ConcludingRem","Reliability","ReliRem","General
Rem","RefRem","Completed"
15022002093307.0,4,3/10/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: PET 700811
Purity: Assume 100% for dose calculations (actual, 99.7%)
Additions: None reported.
Solvent Carrier: Olive oil (0.55 ml/kg)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.3100","Yes",1983,"
rat","Fischer 344","Both",20,20,"Oral",94,"once daily for 94 days"
,"0, 100, 300, and 900 mg/kg/day","Yes","Animals were sacrificed a
fter last dose.","Two-tailed Student's t-test","This study was de
signed to evaluate the toxic effects of PET when administered dail
y by oral gavage to rats for 13 weeks and to determine target orga
ns. Male and female Fischer 344 rats (age 40 days) were randomly
assigned to control (olive oil, 0.55 ml/kg), 100, 300, and 900 mg/
kg/day PET (20 males and 20 females per group). Dosing solutions
were prepared on three occasions: for dosing Weeks 1 through 4; fo
r dosing Weeks 5 through 8; and for dosing Weeks 9 through study c
ompletion. Dosing solutions were prepared by adding the correct a
mount of test substance to the correct amount of Pompeian olive oi
l and thoroughly mixing on a magnetic stirrer for 10 to 20 minutes
. The volume (ml/kg) and g of dosing solution for control, 100, 3
00, and 900 mg/kg/day were 0.55, 0
.67, 0.90, and 1.60, respectively, and 0, 15.05, 33.39, and 56.33,
respectively (based on the specific gravity of PET, 0.86 g/ml, an
d olive oil, 0.91 g/ml). Dosing solutions were divided into aliqu
ots and placed into glass containers with a minimum of head space
to lessen evaporation of the test substance. The glass containers
were rinsed in acetone and air dried prior to their use. The ali
quots were then frozen at -10 degrees C until shortly before their
use.

Three aliquots from each of the PET treated groups and one aliquot
from the control group were selected after preparation, frozen at
-10 degrees C, and sent for analytical confirmation. The aliquot
s were allowed to thaw and one ml of each aliquot was analyzed by
a Perkin Elmer 3920B Gas Chromatograph and FID.

Dosing solutions were administered daily, 7 days/week by oral gava
ge; each animal received a volume of dosing solution calculated fr
om its most recent body weight and the dosing factor for that trea
tment group. A stainless steel feeding needle fitted to a 0.25 or

0.5 cc BD syringe was used to administer daily gavage doses. Sin
ce the carrier, olive oil, was a digestible oil that added to the
caloric intake of the animals, each animal received 0.5 g of olive
oil/kg body weight per day.

This study was designed for 91 consecutive days of oral gavage exp
osure to PET. Additional dosing was necessary to accomplish the o
bjectives of the study; however, each terminally sacrificed animal
was necropsied a minimum of 20 hours after the last dose (on Day
94).

Animals were observed twice daily for mortality; at least 7 hours
apart on weekdays and at least 4 hours apart on weekends and holid
ays. Animals not expected to survive to the next observation inte
rval were sacrificed at that time. Each animal was observed in it
s cage at the beginning of each day for the presence of obvious ph
armacotoxic or toxicologic signs.

Each animal was examined for appearance, behavior, reaction to han
dling, reflexes, posture, gait, and body discharges on the first d
ay of each study week. The presence or absence of obvious clinica
l signs were noted at this time.

The body weight of each animal was recorded initially (first day o
f study), then weekly for the duration of the study, and at termin
ation just prior to necropsy (fasting weights taken before termina
l sacrifice). Food consumptions were measured for each animal ove
r a 7-day period.

Clinical pathology analyses were performed during Weeks 5 and 13.
Blood specimens were obtained from the orbital sinus of 10 animal
s/sex/group without anesthesia. The animals were fasted approxima
tely 20 hours before blood collection and the blood samples were c
ollected and analyzed in an order that rotated through the treatme
nt groups and sexes. Blood samples were collected in blood tubes
appropriate for the type of analysis required. Urine samples were
collected in metabolism pan run-off into gauze filtered plastic u
rinalysis cups.

Animals that died were necropsied immediately after being found.
Those animals sacrificed moribund also were necropsied immediately
after euthanasia. In all cases necropsies were performed within
16 hours of death. At the study conclusion, all surviving rats we
ighed, anesthetized by carbon dioxide asphyxiation, exsanguinated,
and necropsied in an order that rotated through the treatment gro
ups and sexes. Each animal received its final dose approximately
20 hours before necropsy. The following organs and tissues were r
emoved following gross pathologic examination and preserved in 10%

neutral buffered formalin: brain (entire), pituitary, spinal cord, rectum, mesenteric and mandibular lymph node, eyes, salivary glands, thyroid, trachea, thymus, esophagus, heart and aorta, spleen, adrenals, pancreas, skin with mammary gland, tongue, head (entire), duodenum, jejunum, ileum, colon, cecum, urinary bladder, testes/epididymides, prostate, seminal vesicle, ovaries, uterus (entire), vagina, sciatic nerve, bone wi
th marrow, lungs, liver, kidneys, stomach, skeletal muscle, and gross lesions.

In addition to terminal body weights, the following absolute organ weights were determined for each terminally sacrificed animal prior to fixation (paired organs or dual-lobed organs were weighed together): lungs, kidneys, adrenals, liver, heart, brain, spleen, gonads, and thymus. The thyroid/parathyroids were weighed following fixation. Relative organ/body weights were determined for each animal.

All tissues collected from all control and 900 mg/kg/day animals and liver and kidneys from 100 and 300 mg/kg/day animals were processed for microscopic examination. Additionally, testes from all male rats from 100 and 300 mg/kg/day were examined microscopically.

All endpoints of the control group were statistically compared to the treated groups of the same sex using a two-tailed Student's t-test at the 5% probability level.","=",100,"mg/kg-bw","Based on survival, body weight losses, clinical pathology parameters, liver and testicular weights, and testicular histopathology.","=",300,"mg/kg-bw","Based on survival, body weight losses, clinical pathology parameters, liver and testicular weights, and testicular histopathology.","All dosing solutions were within 10% of target.","Significant effects on survival, body weight losses, clinical pathology parameters, liver and testicular weights, and testicular histopathology were observed at 300 and 900 mg/kg/day.","Significant effects (p<0.05) on survival, body weight losses, clinical pathology parameters, liver and testicular weights, and testicular histopathology were observed at 300 and 900 mg/kg/day.","Treatment-related mortality was apparent in the 300 and 900 mg/kg/day male and female groups following eight weeks of PET
700811 oral administration. Cumulative mortality (dead/alive) at Week 8 for males exposed to 0, 100, 300, and 900 mg/kg/day was 0/20, 0/20, 8/20, and 4/20, respectively. Cumulative mortality at Week 13 for males was 0/20, 0/20, 8/20, and 8/20, respectively. Cumulative mortality at Week 8 for females was 0/20, 1/20, 4/20, and 13/20, respectively. Cumulative mortality at Week 13 for females was 1/20, 2/20, 7/20, and 18/20, respectively.

Statistically significant decreases ($p<0.05$) in mean body weights were observed for males at 300 and 900 mg/kg/day during Weeks 1 th rough 8 and the decrease continued through Week 13. Mean body wei ght gains at 0, 100, 300, and 900 mg/kg/day for males were 176, 18 2, 145 (significant at $p<0.05$), and 105 g (significant at $p<0.05$), respectively. No significant mean body weights were observed for the treated female groups after Week 4 due to elimination by deat h or moribund sacrifice of those animals with consistently lower b ody weights.

No pattern or consistent trends were noted in food consumption for treated male or female groups. Sporadic instances of significant ly higher or lower than control mean food consumption vaules were noted: males, 300 mg/kg/day, lower than control during Weeks 1 and 3 and higher than control during Week 7; males, 900 mg/kg/day, lo wer than control during Weeks 1, 3, and 4 and higher than control during Weeks 11 and 12; females, 300 mg/kg/day, lower than control during Week 1 and higher than control during Weeks 5, 8, and 11; females, 900 mg/kg/day, higher than control during Weeks 4, 7, 8, 9, and 10.

Results of the hematology data were somewhat equivocal; data from the Weeks 5 and 13 analyses indicated variable erythropoietic chan ges in males at 300 and 900 mg/kg/day, decreased numbers of platel ets in male and female treated groups, and differential leukocytic changes in all treated male and female groups. Erythropoietic ch anges involved statistically significantly elevated mean RBC, HGB, and HCT values in the 300 and 900 mg/kg/day males during Week 5. No evidence of erythropoietic changes were observed in the treate d female groups during the Week 5 interval; a significantly higher than control mean hematocrit level was noted in 900 mg/kg/day but was considered statistically incidental. By Week 13, all indicat ions of hematoconcentration were absent from 300 and 900 mg/kg/day males. Statistically significatly lower than control mean RBC an d HGB levels were noted in 100 mg/kg/day females during Week 13 an d were considered incidental.

Leukocytic changes consisted of statistically significantly elevat ed mean total leukocyte counts (WBC) in the 300 and 900 mg/kg/day male and female groups during Week 5 and an apparent leukocyte dif ferential shift in all treated male and female groups during Weeks 5 and 13 when compared to the control group. As with the erythro poietic changes observed in 300 and 900 mg/kg/day males, elevation s in the WBC counts in the 300 and 900 mg/kg/day males and females were highest during Week 5 with a return to normal levels by Week 13.

A dose-related shift in the leukocytic differential count occurred in all treated male groups and consisted of increases in segmente d neutrophils with decreases in lymphocytes at Weeks 5 and 13. Th e leukocytic shift appeared to be most evident at Week 5 with only minimal abatement by Week 13. In contrast, all treated female gr oups showed increases in segmented neutrophils with corresponding minimal increases in lymphocyte number as well; no dramatic differ ences were observed in the intensity of the differential shift bet ween Weeks 5 and 13.

Absolute leukocyte differential counts were calculated for Week 13 and Week 14. Week 13 absolute leukocyte differential counts were calculated for 10 rats/sex/group. Week 14 absolute leukocyte dif ferential counts were calculated for all surviving animals (in add ition to the 10/sex/group) in order to increase the statistical an d interpretive reliability of the leukocyte differential shift obs erved at Week 13. The percentages of increase or decrease in segm ented neutrophils and lymphocytes in the treated groups compared w ith controls were as follows: (Week 13; 100, 300, and 900 mg/kg/da y, respectively) male segs., +37, +46, +112; male lymphs., -3, -12 , -29; female segs., +3, +21, +119; female lymphs., +9, +4, +18. Week 14, male segs., +35, +56, +128; male lymphs., +1, -20, -27; f emale segs., +4, +18, +128; female lymphs., +7, -1, +10.

Statistically significant elevations in the mean absolute segmente d neutrophils of the 300 and 900 mg/kg/day male groups were noted during Weeks 13 and 14. Concomitantly, statistically significant decreases in the mean absolute lymphocytes were noted for the 900 mg/kg/day males during Week 13 and for 300 and 900 mg/kg/day males during Week 14. These values were within the historical ranges f or control, however the normal range is very broad (# of cells/cum m, males, 4544-10788; females, 3300-9100). When compared to the c ontrol group, the increased numbers of segmented neutrophils for a ll treated male groups with corresponding decreases in total lymph ocytes in the 300 and 900 mg/kg/day males showed a treatment relat ionship. Segmented neutrophil and lymphocyte counts for the 100 a nd 300 mg/kg/day females were similar to those of the control grou p. A statistically significant increase in the mean absolute segm ented neutrophil count was noted for the 900 mg/kg/day females dur ing Weeks 13 and 14. Data for the
900 mg/kg/day females suggested a treatment-related increase in se gmented neutrophils and possibly in lymphocytes. The shift in the differential counts may be related to stress inducement as a resu lt of dosing with PET.

Statistically significantly lower than control mean platelet count s were observed for the 900 mg/kg/day males during Week 5 and for the 300 and 900 mg/kg/day males during Week 13. In addition, thes e values were lower than historical controls. Mean platelet count

s of the 100 mg/kg/day males were numerically lower than control v
alues during Weeks 5 and 13. Statistically significantly lower th
an control mean platelet counts were observed for the 100 and 300
mg/kg/day females during Week 13 and numerical decreases in mean p
latelet counts were noted in all other treated female groups at bo
th intervals (except 100 mg/kg/day, Week 5). Decreases in platele
t cell number appeared to be dose-related in both male and female
treated groups by Week 13.

Results of the clinical chemistry analyses showed treatment-relate
d statistically significant increases in alanine aminotransferase
(SGPT), alkaline phosphatase (ALP), and albumin levels in the male
 and female 300 and 900 mg/kg/day groups during Week 5. Total pro
tein levels for 900 mg/kg/day males and females were also elevated
 at this interval. Treatment-related statistically significant re
ductions in total cholesterol (300 and 900 mg/kg/day males and 900
 mg/kg/day females) and glucose levels (900 mg/kg/day males) also
were noted during Week 5. Statistically significantly elevated ca
lcium levels for male and female 900 mg/kg/day groups noted at Wee
k 5 were within normal limits for rats of this age and strain and
were not considered treatment-related.

By Week 13, treatment-related increases in SGPT, ALP, total protei
n, and albumin levels were confined to the 900 mg/kg/day males and
 females only and were no longer observed in the 300 mg/kg/day mal
es and females (except statistically elevated ALP in the 300 mg/kg
/day females). Treatment-related statistically significant reduct
ions in total cholesterol and glucose levels were sustained in the
 900 mg/kg/day males during Week 13 and also present in the 300 mg
/kg/day males.

Additional noteworthy statistically significant differences betwee
n control and treated clinical chemistry data were observed. The
following were considered to be incidental to treatment: higher t
han control mean potassium value for the 100 mg/kg/day males durin
g Week 5 and higher than control mean albumin value during Week 13
; and higher than control mean albumin/globulin ratio and lower th
an control mean sodium value for the 300 mg/kg/day females during
Week 13.

Results of the urinalyses were unremarkable.

No consistent treatment-related trends were evident in the necrops
y data from animals found dead and sacrificed moribund or from ani
mals sacrificed by design at the conclusion of the study. Gross l
esions observed with comparable incidence between control and trea
ted groups were characterized as those normally occurring in rats.
  The incidence of brain lesions (meninges reddened, swelling of t

he brain or red streaking) was somewhat higher in 900 mg/kg/day fe males which died or were sacrificed moribund; however, no correspo nding lesions were evident microscopically.

A variety of noteworthy organ/body weight data was noted; however, dose-related findings were confined to increases in absolute and relative liver weights of all treated male and female groups and r eductions in absolute and relative testes/epididymides of the 300 and 900 mg/kg/day males. Statistically significantly or numerical ly higher absolute and relative liver weights were noted for all t reated male and female groups. Relative liver weights for 100 mg/ kg/day males and females were 3.9% and 6.4% higher than their resp ective control group. These slight increases were considered inci dental to treatment. Absolute testes/epididymides weights of the 300 and 900 mg/kg/day males were statistically lower than the cont rol absolute weight; only the relative testes/epididymides weight of the 900 mg/kg/day males was significantly lower than control. Mean absolute liver weights for males at 0, 100, 300, and 900 mg/k g/day, respectively, were 8.08, 8.28, 8.53, and 9.68 g (significan t at $p<0.05$). Mean relative liver
weights for males were 2.97, 3.08 (significant at $p<0.05$), 3.58 (s ignificant at $p<0.05$), and 4.89% (significant at $p<0.05$). Mean ab solute liver weights for females were 4.66, 4.88 (significant at p <0.05), 5.13 (signficant at $p<0.05$), and 6.95 g (significant at p< 0.05). Mean relative liver weights for females were 2.93, 3.11 (s ignificant at $p<0.05$), 3.39 (significant at $p<0.05$), and 4.86% (si gnificant at $p<0.05$). Mean absolute testis/epididymides weights f or males were 4.06, 4.12, 3.80 (significant at $p<0.05$), and 1.85 g (significant at $p<0.05$). Mean relative testis/epididymides weig hts for males were 1.51, 1.56, 1.60, and 0.931% (significant at p< 0.05).

The following statistically significant organ weight data were con sidered to be incidental as differences between control and treate d values were a direct result of significantly lower body weight f or the 300 and 900 mg/kg/day groups at the time necropsy or were a result of purely incidental statistical occurrences: lung, relati ve, 300 mg/kg/day males, higher than controls; lung, absolute/rela tive, 900 mg/kg/day males; lower/higher than controls; kidney, abs olute, 300 mg/kg/day males, lower than control; kidney, absolute/r elative, 900 mg/kg/day males, lower/higher than controls; heart, r elative, 300 and 900 mg/kg/day males, higher than controls; thyroi d/para, relative, 100 and 900 mg/kg/day males, higher than control ; brain, relative, 300 mg/kg/day males, higher than control; brain , absolute/relative, 900 mg/kg/day males, lower/higher than contro l; spleen, relative, 300 mg/kg/day males, higher than controls; sp leen, absolute/relative, 900 mg/kg/day males, lower/higher than co ntrols; lung, relative, 300 mg/kg/

day females, higher than controls; kidney, relative, 300 and 900 m g/kg/day females, higher than controls; adrenal, absolute/relative , 300 mg/kg/day females, higher than controls; heart, absolute, 90 0 mg/kg/day females, lower than control.

The pathology findings were as follows: Fourteen of the twenty 900 mg/kg/day males exhibited testicular atrophy and decreased sperma togenesis. Sperm were also decreased or absent from the epididymi des in these animals. A single control rat had testicular atrophy and the accompanying aspermatogenesis and aspermia. Eight of the 900 mg/kg/day rats that had atrophic testicles also had sperm gra nulomas in the epididymides; four also had atrophy of the seminal vesicles. The atrophic genital organs may have been associated wi th malnutrition and weight loss; the relationship of sperm granulo mas to any of these changes is unknown. Sections of testicles fro m all of the male rats in 100 mg/kg/day appeared normal. There wa s no microscopic indications of atrophy in the sections of testicl es from 300 mg/kg/day rats; however, two of these animals had mini mal hypospermatogenesis. This change was characterized by occasio nal tubules having a decrease in spermatids in the tubule lining c ells and no mature sperm in the lu
mens. The change was of such a slight degree there was no apparen t decrease in the bulk of the testicle.

There were no microscopic changes in the ovaries or other organs o f the female reproductive system that were comparable to those in the males. Few other inflammatory, degenerative, or developmental changes were present in the remaining tissues. Inflammatory chan ges of the eye and its adnexa, lung, and kidney were present in bo th males and females in the control and 900 mg/kg/day animals with about equal frequency. A few examples of centrilobular hepatocyt omegaly were seen in the livers of both test and control rats. Th ere were rare ovarian cysts and distended uteri in the females. T he only findings in the gastrointestinal tract were nematodiasis i n the colon and single examples of bile staining in the stomach an d submucosal hemorrhages in the jejunum.","Three groups of Fischer 344 rats received oral gavage doses of PET 700811 at 100, 300, an d 900 mg/kg/day for 13 consecutive weeks to evalute the toxic effe cts and to determine target organs. Dose-related mortality was ob served for the 300 and 900 mg/kg/d
ay males and females. Significant body weight depression (p<0.05) and lowered body weight gains were noted for the 300 and 900 mg/k g/day males throughout the course of the study. Body weights of t he 300 and 900 mg/kg/day females were significantly lower than con trols during the first four weeks of study. Noteworthy difference s in food consumption between control and treated groups were not observed. No treatment-related pharmacotoxic signs were observed in either sex at any dose level.

Hematology data indicated hematoconcentration in the male 300 and 900 mg/kg/day groups during Week 5 with complete abatement by Week 13. No similar findings were observed in female treated groups. A leukocyte differential shift was evident in all treated male an d female groups during Weeks 5, 13, and 14 and consisted of increa sed numbers of segmented neutrophils with corresponding decreases in lymphocyte numbers in the treated male groups and increased num bers of both segmented neutrophils and lymphocytes in the treated female groups; the shift appeared to be treatment related in the 3 00 and 900 mg/kg/day females. Dose-related reductions in platelet cell number were observed in all treated male and female groups b y Week 13.

Treatment-related elevations in SGPT, ALP, and albumin levels were present in the male and female 300 and 900 mg/kg/day groups durin g Week 5, as well as significant reductions in total cholesterol l evels (300 and 900 mg/kg/day males and 900 mg/kg/day females) and glucose levels (900 mg/kg/day males). Total protein levels for th e 900 mg/kg/day males and females were also elevated at Week 5. T reatment-related elevations in SGPT, ALP, total protein, and album in levels were confined to the 900 mg/kg/day males and females dur ing Week 13, as well as treatment-related reductions in total chol esterol and glucose levels in the 300 and 900 mg/kg/day males.

Results of the urinalysis were unremarkable. No consistent treatm ent-related trends in gross necropsy data were evident. The absol ute and relative liver weights of 300 and 900 mg/kg/day male and f emale groups increased and a dose-related pattern was observed. N o corresponding liver lesions were seen upon histopathological eva luation.

Dose-related reductions in absolute and relative testes/epididymid es weights of the 300 and 900 mg/kg/day males were observed. Micr oscopically, administration of PET was associated with testicular atrophy and hypospermatogenesis of the testes and hypospermia or a spermia of the epididymides in the 900 mg/kg/day males; a number o f these animals had sperm granulomas in the epididymides. No micr oscopic indication of atrophy was seen in the sections of testicle s from the 300 mg/kg/day rats; however, two of the animals showed minimal hypospermatogenesis. Testicle sections from all 100 mg/kg /day males appeared normal.

Based on the results, the NOAEL and LOAEL were 100 and 300 mg/kg/d ay, respectively. Definitive effects were observed on survival, b ody weight losses, clinical pathology parameters, liver and testic ular weights, and testicular histopathology at both 300 and 900 mg /kg/day.","Acceptable","All key parameters (i.e., number of animal

s, doses, observations, etc.) were appropriate and adequately desc
ribed in the study.",,"Thirteen Week Gavage Administration of PET
(MEHSL Sample No. 700811) to Rats. Borriston Project No. 3401. B
orriston Laboratories, Temple Hills, MD (701-81).","Y"
15022002093307.0,1,3/7/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-143-79
Purity: Assume 100% for dose calculations (actual, 95.6%)
Additions: Toluene, ethylbenzene, p-xylene, cymenes and m-ethyltol
uene (4.4%)
Solvent Carrier: None reported
Contaminants: None reported
Chemical formula: C9H12",,"Other","Yes",1979,"rabbit","New Zealand
white","Both",10,10,"Dermal",14,"Daily","0, 200, 500, 1000, and 2
000 mg/kg","Yes","Animals were sacrificed after 14-days of dosing.
","ANOVA with Scheffe's multiple pairwise comparisons","The purpos
e of this study was to evaluate the systemic and dermal effects in
rabbits resulting from repeated dermal contact with MCTR-143-79 f
or fourteen consecutive days. New Zealand White rabbits (initial
body weights of males ranged from 2,953 to 3,425 g and initial bod
y weights of females ranged from 3,112 to 3,400 g) were used in th
e study. Two rabbits of each sex were assigned to the following t
reatment groups: 0, 200, 500, 1000, and 2000 mg/kg. Prior to test
initiation, the hair was closely clipped from the back of each ra
bbit. Just prior to compound application and again on Day 7, the
skin of one male and one female in each group was abraded with min
or incisions sufficiently deep enough to penetrate the stratum cor
neum, but not deep enough to distu
rb the derma or to produce bleeding. The skin of the remaining rab
bits was left intact. The appropriate amount of test substance wa
s applied daily by gentle inunction to the skin of each rabbit for
fourteen consecutive days. The test substance was not wiped off
at any time during the fourteen-day exposure. Rabbits were fitted
with plexiglass collars which were worn for the duration of the s
tudy. The test substance was administered dermally because the po
tential human exposure is by the dermal route.

All of the rabbits were observed twice daily for mortality and onc
e daily for signs of toxic and pharmacologic effects for fourteen
consecutive days. Dermal responses were graded and scored daily i
mmediately prior to the next application according to the system o
f Draize (1959, Appraisal of the Safety of Chemicals in Foods, Dru
gs and Cosmetics, 46-59). A key used in scoring erythema and edem
a is as follows:
Erythema and eschar formation:
0=no erythema
1=very slight erythema (barely perceptible)
2=well-defined erythema
3=moderate to severe erythema

4=severe erythema (beet redness) to slight eschar formation (injur ies in depth)

Edema formation:
0=no edema
1=very slight edema (barely perceptible)
2=slight edema (edges of area well defined by definite raising)
3=moderate edema (raised approximately 1.0 mm)
4=severe edema (raised more than 1.0 mm extending beyond the area of exposure)

Individual body weights were recorded prior to treatment, on Day 7 , and at termination. At termination (day 14), all rabbits were s acrificed with sodium pentobarbital and necropsied. The terminal body weight, body weight change, food consumption, and organ weigh t data of the control group were compared statistically to data of the treated groups of the same sex by Bartlett's test for homogen eity of variance and the one-way analysis of variance (ANOVA). If significant results were obtained from both Bartlett's test and A NOVA, a multiple pairwise comparison procedure was used to compare the group mean values. If a significant result was not obtained from Bartlett's test, but was obtained from ANOVA, Scheffe's multi ple pairwise comparison procedure was used to compare the group me an values. All analyses were evaluated at the 5.0% probability le vel.","<",200,"mg/kg-bw","Skin reaction - erythema (slight), thick ening, fissuring, and necrosis.","<",200,"mg/kg-bw","Skin reaction - erythema (slight), thickening,
fissuring, and necrosis.","Not analytically measured.","No mortali ty was observed at any treatment group. At termination, all anima ls were observed to have thickening, fissuring, and necrosis. Bod y weights were lower at 2000 mg/kg in males and 1000 and 2000 mg/k g in females.","In males, body weights at 14 days were significant ly decreased at 2000 mg/kg (p<0.05). In females, body weight chan ge was significantly decreased at 1000 and 2000 mg/kg (p<0.05).","No mortality was observed throughout the fourteen-day observation period. Three rabbits in control, all rabbits in 200, 500, and 10 00 mg/kg, and one rabbit in 2000 mg/kg appeared normal with respec t to clinical observations throughout the two-week period. Marked anorexia was noted in one control male and one 2000 mg/kg female on Day 3. Slight ataxia (Days 13 and 14) and slight depression (D ay 14) were noted in one female rabbit at 2,000 mg/kg. Thinness a nd eye discharge (Days 11 through 14) were well as slight depressi on and slight ataxia (days 13 and
14) were observed in one male rabbit at 2,000 mg/kg.

No edema was noted in any of the rabbits. Erythema was not noted in any control animals, but was present in treated groups ranging from very slight to well-defined. Three animals in 200 mg/kg, one

in 500 mg/kg, two in 1000 mg/kg, and one in 2000 mg/kg were obser ved to have only very slight erythema, while the remaining animals exhibited erythema ranging from very slight to well-defined. The re appeared to be no difference in erythema between male and femal e rabbits or between abraded and intact skin. Other dermal effect s generally began on Day 3 or 4 with thickening and progressed to include blanching, fissuring, fissuring with bleeding, sloughing, raw areas, and necrosis. Individual scores at day 14 were as foll ows (male abraided, intact, followed by female abraided, intact, r espectively): controls, 0,0,0,0; 200 mg/kg, 1,1,1,1; 500 mg/kg, 1, 2,2,2; 1000 mg/kg, 1,1,2,2; 2000 mg/kg, 1,2,1,1. The individual d ermal irritation scores (T=thickening, F=fissuring, N=necrosis, S= sloughing, R=raw areas) were as fo
llows: Controls, normal; 200 mg/kg, TFN, TFN, TFN, TFN; 500 mg/kg, TFN, TFNS, TFNSR, TFNSR; 1000 mg/kg, TFNR, TFNSR, TFNSR, TFNS; 20 00 mg/kg, TFNSR, TFNSR, TFNS, TFNS.

Mean and individual body weights for the treated male and female g roups were lower than the respective control group. The mean term inal body weight for 2000 mg/kg males was significantly lower than controls. There was a dose-related decrease in mean body weight for the female treated groups at both 7 and 14 days. With the exc eption of 200 mg/kg males gaining signficantly more weight than co ntrols, the body weight change (Days 0-14) followed a dose-related pattern in both male and female groups with males at 500, 1000, a nd 2000 mg/kg losing weight and females in 1000 and 2000 mg/kg sig nificantly losing weight. Mean body weight at 14 days for males a t control, 200, 500, 1000, and 2000 mg/kg was 3427, 3358, 3174, 32 18, and 2506 g, respectively (significant at $p<0.05$). Body weight change for males at control, 200, 500, 1000, and 2000 mg/kg was 1 02, 195 (significant at $p<0.05$), -7.0, -146, and -625 g, respectiv ely. Mean body weight at 14 days for females at control, 200, 500 , 1000, and 2000 mg/kg was 3614, 3
379, 3268, 2970, and 2728 g, respectively. Body weight change for females at control, 200, 500, 1000, and 2000 mg/kg was 294, 215, 44, -282 (significant at $p<0.05$), and -447 g (significant at $p<0.05$), respectively.

No significant differences were noted in the total food consumptio n values. There appears, however, to be a dose-related decrease i n the Day 14 and total food consumption values for the females, wh ile no similar pattern was apparent for the males.

Statistical analyses revealed significantly lower than control val ues for absolute and relative (kidney/body weight ratio) kidney we ights in females at 200 mg/kg and a significantly higher than cont rol value for the mean relative kidney weight in females at 2000 m g/kg. There appeared to be a dose-related decrease in absolute an d relative liver weights in the females. Due to the small sample size, it is difficult to determine the biological significance of these results.

Gross pathology findings in all treated groups consisted of the sk
in lesions of erythema, necrosis, thickening, and fissuring. Inci
dental findings consisted of an enlarged thyroid (control female)
and gallbladder (male, 2000 mg/kg) and a small amount of body fat
(male, 2000 mg/kg).","MCTR-143-79 was evaluated in a fourteen-day
repeated dermal application study in rabbits at dose levels of 0,
200, 500, 1000, and 2000 mg/kg. No rabbits were found dead throug
hout the 14-day observation period. Erythema was noted in treated
 rabbits ranging from very slight to well-defined. Thickening, bl
anching, fissuring (with and without bleeding), necrosis, sloughin
g, and raw areas were noted among treated rabbits. As a result, t
he NOAEL was below 200 mg/kg.","Acceptable","All key parameters (i
.e., doses, observations, etc.) were appropriate and adequately de
scribed.",,"Fourteen-Day Dermal Pilot Study in Rabbits, MCTR-143-7
9. Hazleton Laboratories America, Inc., Vienna, Virginia, Februar
y 15, 1980 (M1430-79).","Y"
15022002093307.0,2,3/8/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: PET
Purity: >99.7% (100% used for dose calculations)
Additions: None reported
Solvent Carrier: Olive oil (10 ml/kg)
Contaminants: None reported
Chemical formula: C9H12",,"Other","Yes",1982,"rat","Fischer 344","
Both",5,5,"Oral",14,"Once daily.","0, 25, 50, 100, 200, 400, and 8
00 mg/kg/day","Yes","Animals were sacrificed after 14 days of dosi
ng.","Bartlett's, ANOVA, Scheffe's multiple comparison","The purpo
se of this study was to evaluate the toxic effects of PET when adm
inistered daily by oral gavage to rats for two weeks and to provid
e data for dose selection for a thirteen-week study. Four-week ol
d Fischer 344 rats were acclimated to laboratory conditions for fi
ve days and 5 male and 5 female rats were each assigned to the fol
lowing concentrations by a HLA weight randomization program: 0 (ol
ive oil, 1.1 ml/kg), 25, 50, 100, 200, 400, and 800 mg/kg/day. Fi
ve male and female rats at each concentration were dosed by oral g
avage daily for 14 days. Ten male and female rats were used for t
he controls. The HLA weight randomization program randomly distri
buted the animals into the desired number of groups and analyzed i
ndividual body weight data to insu
re lack of significant differences between group mean body weights
 at initiation of treatment.

Appropriate amounts of the vehicle and test substance were weighed
 into beakers and mixed by stirring for five minutes with a magnet
ic stirrer. Equal aliquots of each dosing solution were transferr

ed into 30 ml amber vials and frozen until just before use. Three 10 ml aliquots were verified analytically using a Varian Cary 219 spectrophotometer (273.8 nm).

All animals were observed daily for signs of abnormal appearance, behavior, excretory function, and discharges. Animals were checke d twice daily for mortality and moribundity. Body weights, food c onsumption, and abnormal clinical signs were recorded weekly. All animals were fasted in metabolic cages for at least sixteen hours prior to blood collection. Immediately prior to sacrifice, blood samples were obtained for hematology and serum clinical chemisty by orbital sinus puncture, and urine was collected overnight from the metabolic cages for urinalysis endpoints.

At termination (following blood collection), all animals were sacr ificed with sodium pentobarbital, exsanguinated, and necropsied in an order which rotated through the treatment groups. Organs were weighed and preserved in 10% neutral buffered formalin for histop athologic evaluation.

Mean body weights at Weeks 1 and 2, total food consumption, and cl inical laboratory and organ weight data of the control group were compared statistically to the data of the treated groups of the sa me sex by Bartlett's test for homogeneity of variance. This was f ollowed by ANOVA. Nonparametric data was analyzed by either Schef fe's multiple pairwise comparison or Games and Howell's multiple p airwise comparison. All analyses were evaluated at the 5.0% (one-tailed) level.","=",200,"mg/kg-bw","Increase in male and female me an relative and absolute liver weights at 400 and 800 mg/kg/day.", "=",400,"mg/kg-bw","Increase in male and female mean relative and absolute liver weights at 400 and 800 mg/kg/day.","All dosing solu tions were within 10% of nominal.","No mortality was observed at a ny dose level. An increase in male and female mean relative and a bsolute liver weights was observed at 400 and 800 mg/kg/day. The m ean terminal body weight of males at 800 mg/kg/day was significant ly lower than controls. A signifi
cantly higher than control mean serum glutamic pyruvic transaminas e value was observed for males at 800 mg/kg/day. Most frequently noted gross pathology findings were discolored thymus or lungs; re ddened, thin, or smoth glandular portion of the stomach; reddened or enlarged lymph nodes; and distended uterine horns.","A statisti cally significant increase (p<0.05) in male and female mean relati ve and absolute liver weights at 400 and 800 mg/kg/day.","No death s occurred during the study. All animals appeared normal througho ut the study except one female at 50 and 100 mg/kg/day. The femal e at 50 mg/kg/day appeared hunched and thin with urine stains, rou gh haircoat, and bloody, crusted eyes at one observation during We ek 2. Urine stains were noted for the female at 100 mg/kg/day dur

ing Week 2.

Statistical comparison of mean body weights of the treated groups with the control group for each sex revealed a significantly lower mean body weight for females at 800 mg/kg/day during Week 1. Mea n body weights for all other treated groups were comparable to the respective control group. Mean body weights for females at 0, 25 , 50, 100, 200, 400, and 800 mg/kg/day (Week 1) were 109, 105, 101 , 107, 108, 106, and 99 g (significant at $p<0.05$), respectively. Mean body weights for females (Week 2) were 122, 115, 112, 123, 12 4, 122, and 110 g, respectively. Mean terminal body weights for f emales were 111, 106, 102, 112, 113, 112, and 99 g, respectively. The mean terminal body weight of males at 800 mg/kg/day was signi ficantly lower than control. Mean body weights for males at 0, 25 , 50, 100, 200, 400, and 800 mg/kg/day (Week 1) were 150, 150, 148 , 152, 148, 143, and 134 g, respectively. Mean body weights for m ales (Week 2) were 178, 176, 177, 182, 178, 171, and 158 g, respec tively. Mean terminal body weight
s for males were 161, 159, 157, 163, 161, 152, and 138 g (signific ant at $p<0.05$), respectively.

Total food consumption for females at 800 mg/kg/day was significan tly less than the control group, but all other treated groups were comparable to the control group of the same sex. No significant differences were observed in males. Mean food consumption for mal es at 0, 25, 50, 100, 200, 400, and 800 mg/kg/day (Week 2) was 115 , 109, 111, 115, 113, 111, and 107 g, respectively. Mean food con sumption for females (Week 2) was 84, 80, 73, 87, 91, 87, and 79 g (significant at $p<0.05$), respectively.

No treatment-related trends were evident in the clinical laborator y data; however, statistical evaluation revealed a significantly h igher than control mean serum glutamic pyruvic transaminase value for males at 800 mg/kg/day. The mean alkaline phosphatase value f or females at 50 mg/kg/day was higher than other treated groups an d the control group of the same sex. The large standard deviation reflected elevated alkaline phosphatase in a single animal which was not believed to be treatment related. All other hematology an d clinical chemistry values were comparable to the respective cont rols and were within normal ranges for this age and species of ani mals. Urinalysis findings were unremarkable.

Mean relative and absolute liver weights were significantly higher than control values for males and females at 400 and 800 mg/kg/da y. Liver weights for males at 0, 25, 50, 100, 200, 400, and 800 m g/kg/day were 5.5, 5.4, 5.5, 5.8, 5.8, 5.9, and 6.4 g (significant at $p<0.05$), respectively. Liver ratios for males were 3.4, 3.4, 3.5, 3.5, 3.6, 3.9 (significant at $p<0.05$), and 4.7% (significant

at p<0.05), respectively. Female liver weights were 4.0, 3.8, 3.8 , 4.0, 4.3, 4.5 (significant a p<0.05), and 4.4 g, respectively. Female liver ratios were 3.6, 3.6, 3.7, 3.6, 3.8, 4.0 (significant at p<0.05), and 4.4% (significant at p<0.05), respectively. Mean relative kidney weight for males increased with increasing dose w ith the 800 mg/kg/day value being significantly elevated. However , a similar dose-related trend was not observed in the absolute ki dney weights for the males or in the absolute or relative kidney w eights for the females. The mean relative adrenal weights for mal es at 800 mg/kg/day was significan
tly greater than control, but this was considered attributable to the low male mean terminal body weight.

Most frequently noted gross pathology findings were discolored thy mus or lungs; reddened, thin, or smooth glandular portion of the s tomach; reddened or enlarged lymph nodes; and distended uterine ho rns. Cysts on the pituitary, discolored areas or nodules on the l ung, discolored cecum, and nodules on the liver adhered to the dia phragm were also found.

The analyses of PET dosing solutions yielded the following % from nominal for 25, 50, 100, 200, 400, and 800 mg/kg/day: 105, 96, 97, 97, 98, and 97, respectively.","A two-week oral gavage study of P ET in male and female rats resulted in a NOAEL and LOAEL of 200 an d 400 mg/kg/day, respectively, based on an increase in male and fe male mean relative and absolute liver weights. No mortality was o bserved at any treatment group during the study. Lower than contr ol mean body weights were noted for 400 and 800 mg/kg/day animals with statistically significant decreases at termination for the ma les (800 mg/kg/day) and at Week 1 for the females (800 mg/kg/day). Some apparently treatment-related differences were also noted in the absolute and relative organ weight data for 400 and 800 mg/kg /day. Mean relative liver weights were significantly higher than control for 400 and 800 mg/kg/day animals of both sexes with great er mean absolute liver weights for the 400 mg/kg/day females and 8 00 mg/kg/day males. The mean rela
tive kidney weight for 800 mg/kg/day males was significantly highe r than control and mean values for the other treated groups showed an apparently dose-related trend. However, this trend was not re flected in the female data. Clinical laboratory data and gross pa thology findings were considered unremarkable.","Acceptable","All key parameters (i.e., doses, observations, etc.) were appropriate and adequately described in the study.",,"Two-Week Gavage Administ ration of PET to Rats. Project No.: 230-235. Hazleton Laboratorie s America, Inc., February 9, 1982 (702-81).","Y"
15022002093307.0,3,3/9/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-51-79
Purity: Assume 100% for dose calculations (actual, 95.6%)

Additions: Toluene, ethylbenzene, p-xylene, cymenes and m-ethyltol
uene (4.4%)
Solvent Carrier: None
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.3465","Yes",1979,"
rat","Fischer 344","Both",15,15,"Inhalation",90,"6hrs/day, 5 days/
week for 13 weeks","0, 100, 300, and 1000 ppm","Yes","All animals
sacrificed after 13 weeks exposure.","ANOVA, Bartlett's, Dunnett's
, Kruskal-Wallis, Dunn","The purpose of this study was to assess t
he toxic effects of MCTR-51-79 when administered to rats, six hour
s per day, five days per week, for 13 weeks at target concentratio
ns of 100, 300, and 1000 ppm. Male and female Fischer 344 (CDF) r
ats (48 days old; males ranged from 121-153 g and females ranged f
rom 90 to 119 g) were used. Animals were observed twice daily dur
ing each exposure and nonexposure day for abnormal signs and a ful
l, recorded, physical assessment was performed weekly. Individual
body weights were recorded 7 and 4 days prior to exposure, Day 1,
Day 2, and weekly thereafter through termination. Hematology, cl
inical chemistry, and urinalysis parameters were measured on a ran
domly selected group of five anima
ls per sex during Week 5 and on all survivors following the 13 wee
k exposure. Ophthalmoscopic examinations were performed on all an
imals prior to and following the 13 week exposure. Gross necropsy
examinations were performed on all survivors following the 13 wee
k exposure and microscopic examinations were performed on tissues
from all control and 1000 ppm animals.

For 100 and 300 ppm exposures the test substance was placed in a 8
00 ml gas washing bottle and then placed in a waterbath maintained
at 60 degrees C by a Braun Thermomix. For the 1000 ppm exposure
group the test substance was placed in a 1,000 ml gas washing bott
le and then placed in a waterbath maintained at 80 degrees C by a
magnetic stirrer with hotplate and temperature probe. Dry air, at
various flow rates, was passed through the test substance in each
bubbler to create a vapor. The resultant vapor-laden airstream f
rom each bubbler passed through glass tubing and into the entry po
rtal of a one cubic meter stainless steel and glass exposure chamb
er. Chamber air flow was maintained at 173 liters per minute in a
ll exposure chambers. The stainless steel and glass chambers in w
hich the animals were exposed had a total volume of one cubic mete
r with an effective exposure volume of one cubic meter. They were
operated dynamically at an air flow rate of 173 liters per minute
. This flow rate provided one com
plete air change every 5.8 minutes and a 99% equlibrium time of 26
.7 minutes.

Three air samples were drawn from each exposure chamber using a Wi
lks Instrument Company, Miran Long Pathlength Infrared Analyzer Mo

del IA on each exposure day. Samples were drawn at approximately the first, third, and fifth hour of exposure. The concentration of the test material was determined by comparing the absorption of t hese samples to a calibration curve prepared using the same instru mental settings.

Statistical evaluation of equality of means was made by the approp riate ANOVA, followed by a Dunnett's multiple comparison procedure . The Bartlett's test was done to determine if groups had equal v ariance. For nonparametric procedures, the Kruskal-Wallis test wa s used followed by a summed rank test (Dunn) if differences were i ndicated. A statistical test for trend was performed using either standard regression techniques (equal variance) or a Jonckheere's test for nonparametric data. The test for equal variance (Bartle tt's) was conducted at the 1%, two-sided risk level. All other st atistical tests were conducted at the 5% and 1%, two-sided risk le vels.","=",300,"ppm(air)","Increase in absolute and relative liver weight in males and an increase in relative liver weights in fema les at 1000 ppm.","=",1000,"ppm(air)","Increase in absolute and re lative liver weight in males and an increase in relative liver wei ghts in females at 1000 ppm.","0, 104, 305, and 979 ppm.","Increas e in absolute and relative liver w
eight in males and an increase in relative liver weights in female s at 1000 ppm.","Increase in absolute and relative liver weight in males and an increase in relative liver weights in females at 100 0 ppm (p<0.01).","The cumulative mean exposure concentrations for 100, 300, and 1000 ppm nominal concentrations were 104, 305, and 9 79 ppm, respectively, based on measurements made using the Miran M odel IA Infrared Analyzer. On one day during Week 5, one female r at from 300 ppm was not exposed to the test substance. The animal was inadvertently not loaded into the chamber due to a technician error. One animal was mistakenly sorted and placed in the 100 pp m group after pre-test ophthalmoscopy revealed it to have synechia . The animal was treated for the entire 13 weeks of the study.

All animals survived the duration of the 13 week exposure. Howeve r, one female rat from 1000 ppm died accidentally during the Week 5 eye bleeding and two more female animals (one from control and o ne from 1000 ppm) died accidentally during the terminal eye bleedi ng. These deaths were not attributed to the treatment.

Dry rales, excessive lacrimation, dried material around eyes, nos e, or mouth, and yellow staining of the ano-genital fur were obser ved in treated and control groups. Hair loss was noted in all tre ated groups. The higher incidence of yellow staining of the ano-ge nital fur in all treated groups appeared to be exposure related al though a dose-relationship was not statistically confirmed. Hair loss also appeared to be treatment-related in 1000 ppm. All other observations were scattered in appearance and not indicative of a response to the exposure.

Body weights for both male and female animals were unremarkable th roughout the duration of the study. Hematology parameters evaluat ed during Week 5 were unremarkable. During the Week 13 hematology analysis, females at 1000 ppm exhibited a significantly (p<0.01) increased white blood cell level when compared to the control grou p. For example, mean WBC for control and 1000 ppm were 9.2 and 11 .8 1E+3/mm3, respectively. The elevated value, however, fell with in normal biological limits and was not considered to be of biolog ical significance. All other hematology parameters were unremarka ble for both sexes.

During the Week 5 clinical chemistry examination, serum glutamic p yruvic transaminase levels were significantly depressed (p<0.05) i n males at 300 ppm, and alkaline phosphatase levels were significa ntly increased (p<0.01) in males at 100 and 1000 ppm. Clinical ch emistry parameters evaluated for female animals were unremarkable when compared to controls. During the Week 13 clinical examinatio n, serum glutamic pyruvic transaminase levels were significantly d epressed (p<0.01) in 300 and 1000 ppm males. Alkaline phosphatase levels were significantly increased (p<0.01) in 1000 ppm females. All other clinical chemistry parameters for the exposed rats wer e unremarkable in both sexes when compared to controls. The depre ssed serum glutamic transaminase levels in 300 and 1000 ppm males appeared to follow a dose related pattern, but the absolute differ ences were small, within normal biological limits, and the directi on of the effect does not indicate a toxic response. The elevatio n of serum alkaline phosphatase le
vels observed in 100 and 1000 ppm males during Week 5 was not seen at termination, and the absence of a response in 300 ppm implies that this may be a random occurance and unrelated to exposure. Th e elevated serum alkaline phosphatase levels in 1000 ppm females d uring Week 13 does not appear to be biologically significant. Wee k 5 and Week 13 (terminal) urinalysis examinations revealed small amounts of protein in the control group as well as all test groups in both sexes. All other parameters examined for both sexes were comparable to the control.

Statistical analysis of absolute organ weights revealed a signific ant depression (p<0.05) of gonad weights in 1000 ppm males, and a significant (p<0.01) increase in absolute liver weights in 1000 pp m males. All other absolute organ weights were unremarkable when compared to the control. Statistical analysis of organ/body weigh t ratios revealed a significant depression (p<0.05) in the relativ e left kidney weights of 300 ppm males. Relative liver weights we re significantly increased (p<0.01) in the 1000 ppm males and fema

les. All other relative organ weights were unremarkable when comp ared to the control. The increased absolute liver weights in 1000 ppm males and the increased relative liver weights in 1000 ppm ma les and females appeared to be treatment related. The other inter group differences did not indicate a dose-related pattern and did not appear to be of toxicological signficance. Mean absolute live r weights in males for 0, 100, 300, and 1000 ppm were 7.59, 7.52, 8.03, and 8.74 g (significant at p
<0.01), respectively, and mean relative liver weights for males we re 2.72, 2.70, 2.82, and 3.13% (significant at p<0.01), respective ly. Mean absolute liver weights in females for 0, 100, 300, and 1 000 ppm were 4.67, 4.57, 4.59, and 5.25 g, respectively, and mean relative liver weights for females were 2.71, 2.69, 2.73, and 2.83 % (significant at p<0.01), respectively.

There were no evidence of ocular abnormalities in rats exposed to the test substance. Gross and microscopic examinations revealed f ew incidental lesions and tissue changes in both the control and t est animals. There were not considered related to the test substa nce exposures.","An inhalation toxicity study was performed with a vapor of MCTR-51-79. Groups of 15 male and 15 female Sprague-Daw ley rats were exposed 6 hours/day, five days/week, for thirteen we eks. The cumulative mean exposure concentrations were 0, 104, 305 , and 979 ppm.

There were no treatment related mortalities during the study. Obs ervations of the animals during the study showed a higher incidenc e of yellow staining of the ano-genital fur in the exposed animals compared to the controls. This did not follow a dose-response pa ttern but does appear treatment related. Other signs observed dur ing the study were sporatic and did not appear treatment related. Body weights for the exposed animals were comparable to the contr ols and appeared normal.

Hematology findings were all within normal biological limits for e xposed and control rats. Clinical chemistry measurements showed s tatistically significant differences when some of the values for s erum glutamic pyruvic transaminase and serum alkaline phosphatase levels in exposed animals were compared to control levels. None o f these differences were considered to be biologically signficant based on the relatively small differences observed, i.e., all were within physiological limits.

Ophthalmoscopic examinations did not show evidence of exposure rel ated effects.

Analyses of absolute and relative organ weights indicated only one organ, the liver, which may have shown a treatment related respon

se. When compared to the controls, the absolute and relative live
r weights in the 1000 ppm males and females were significantly inc
reased. Gross and microscopic examination of the tissues from the
 control and 1000 ppm rats in this study revealed few incidental l
esions and tissue changes in both the control and test animals. T
hese were not considered related to the test substance exposure.

Based on the increase in liver weights, the NOAEL and LOAEL were 3
05 and 979 ppm, respectively (cumulative mean measured exposure co
ncentrations).","Acceptable","All key parameters (i.e., exposure c
onditions, number of animals, observations, etc.) were appropriate
 and adequately described in the study.",,"A 13 Week Inhalation To
xicity Study of MCTR-51-79 in the Rat. Project No.: 79-7278. Bio
/dynamics Inc., East Millstone, New Jersey, March 31, 1980 (M510-7
9).","Y"

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","TestType","GLP","Year","Species","Strain","Sex","NumberofMales","NumberofFemales","Route","ExposPeriod","Frequency","Doses","ControlGroup","PremExpFemale","PremExpMale","StatMeth","MethodRem","ParNPrec","ParNOEL","ParNUnit","ParNEffect","ParLPrec","ParLOEL","ParLUnit","ParLEffect","F1NPrec","F1NOEL","F1NUnit","F1NEffect","F1LPrec","F1LOEL","F1LUnit","F1LEffect","F2NPrec","F2NOEL","F2NUnit","F2NEffect","F2LPrec","F2LOEL","F2LUnit","F2LEffect","ActualDose","Parental_F1Data","OffspringData","StatResults","ResultsRem","ConcludingRem","Reliability","ReliRem","GeneralRem","RefRem","Completed"

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","TestT
ype","TestSystem","GLP","Year","Species","MetabolicAct","Concentra
tion","StatMeth","MethodRem","Result","CytotoxicConc","GenotoxicEf
f","StatResults","ResultsRem","ConcludingRem","Reliability","ReliR
em","GeneralRem","RefRem","Completed"
15022002093307.0,1,2/21/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-26-79
Purity: 95.6%
Additions: None reported
Solvent Carrier: Dimethylsulfoxide (DMSO, 50 ul)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.5265","Salmonella
typhimurium reverse mutation assay","Bacterial","Unknown",1979,"Sa
lmonella typhimurium","A 9,000 x g supernatant (1 ml) from Sprague
-Dawley adult male rat liver induced by Aroclor 1254","0.0035, 0.0
18, 0.09, 0.18, 0.35 microliters/plate","None referenced","The pur
pose of this study was to evaluate mutagenecity of the test substa
nce in the Salmonella/Mammalian Microsome Pre-Incubation Assay (Ya
hagi, et al. Mutagenicities of N-nitrosamines on Salmonella. Mut
ation Research 48: 121-130, 1977). Top agar was initially prepare
d with 8 g/l Difco Bacto Agar and 5 g/l NaCl. After autoclaving,
100 mls of molten agar was transferred to 100 ml sterile bottles a
nd stored at room temperature. Immediately before use, the top ag
ar was melted and supplemented with 10 ml/100 ml agar of a sterile
 solution containing 0.5 mM L-histidine and 0.5 mM biotin. Bottom
 agar was the Vogel-Bonner minimal medium E described by Ames. Nu
trient broth used for growing over
night cultures of the tester strains contained 25 g/l nutrient bro
th No. 2 (Oxoid). Sham S-9 was the diluent used to adjust the fin
al volume of the non-activation incubation mixture to equal the vo
lume of the activation incubation mixture.

All tester strains (TA98, TA100, TA1535, TA1537, TA1538) were stor
ed in liquid nitrogen, and fresh cultures were inoculated directly
 from these frozen stocks. Broth cultures were grown overnight at
 37degrees C with shaking. In both the toxicity and the mutagenes
is assay, the tester strains were incubated with the test substanc
e for 20 minutes prior to plating. 50 microliters of each tester
strain along with 50 microliters of the appropriate test substance
 dilution was added to glass tubes. The toxicity of the test subs
tance was conducted using the TA100 strain to determine the maximu
m dose to be used in the mutagenesis assay. Concentrations ranged
from 0.0028 to 10 ul/plate and were conducted without metabolic ac
tivation.

In the mutagenesis assay, the test substance was dissolved in DMSO
 and the final concentrations were 0.0035, 0.018, 0.09, 0.18, 0.35
 microliters/plate. In the non-activation assay, 500 microliters

of the Sham S-9 was added to the tester strain-test substance mixt ure. In the metabolic activation assay, 500 microliters of S-9 mi xture (containing the 9,000 x g liver homogenate from adult male r at liver induced by Aroclor 1254) was added to the tester strain-t est substance mixture. After vortexing, the mixtures were allowed to incubate without shaking for 20 minutes at room temperature. The top agar was then added to each tube and the mixture was plate d on Vogel-Bonner bottom agar. The number of cells of each tester strain seeded were as follows: TA98, 1.4E+8; TA100, 1.7E+8; TA153 5, 1.5E+8; TA1537, 0.6E+8; TA1538, 1.3E+8. The plates were incub ated for 48 hrs at 37 degrees C, and scored for the number of reve rtant colonies growing on each plate.

Positive controls were run with each assay. 2-Aminoanthracene (2A A) was pre-incubated and plated at 1.0 ug/plate with metabolic act ivation on strains TA98 and TA100. 2-Nitrofluorene (NF) was pre-i ncubated and plated at 10 ug/plate on strains TA98 and TA1538 with out metabolic activation. Propane sultone (PS) was pre-incubated and plated at 0.4 ul/plate without activation on TA100 and TA1535, and 9-aminoacridine (9AAD) was pre-incubated and plated at 75 ug/ plate on TA1537 without activation. All positive controls, solven t controls, and test substance dilutions were pre-incubated and pl ated in triplicate.

For the test substance to be considered positive, it must cause at least a doubling in the observed revertants per plate of at least one tester strain. The increase in revertants per plate must be accompanied by a dose response to increasing concentrations of the test article.","Negative","The test substance was toxic to the st rain TA100 at 0.29 ul/plate; without metabolic activation","Unconf irmed","All results were negative","The average number of revertan ts per plate for each assay is as follows (all results will be pre sented in the following sequence, solvent, 0.0035, 0.018, 0.09, 0. 18, and 0.35 ul/plate): TA98, Nonactivation, 23, 21, 16, 10, 11, a nd 9; Activation, 39, 33, 34, 30, 24, and 21. TA100, Nonactivatio n, 108, 122, 113, 79, 87, and 69; Activation, 95, 97, 105, 90, 77, and 71. TA1535, Nonactivation, 20, 20, 24, 20, 14, and 0; Activa tion, 19, 25, 23, 20, 11, and 9. TA1537, Nonactivation, 12, 9, 11 , 5, 5, and 0; Activation, 25, 27, 17, 17, 12, and 7. TA1538, Non activation, 16, 15, 22, 15, 12, an
d 0; Activation, 44, 38, 31, 40, 25, and 24. Positive controls we re as follows: TA98, 2AA, activation = 2971; TA98, NF, nonactivati on = 1258; TA100, 2AA, activation = 2589; TA100, PS, nonactivation = 1320; TA1535, PS, nonactivation = 1319; TA1537, 9AAD, nonactiva tion = 572; TA1538, NF, nonactivation = 1128.","The results of the Salmonella/mammalian-microsome pre-incubation mutagenicity assay indicate that the test substance, MCTR-26-79 did not cause a signi ficant increase in the number of revertants per plate of any of th

e tester strains with or without metabolic activation by Aroclor i
nduced rat liver microsomes.","Acceptable","The key parameters (i.
e., dose levels, strains, use of positive controls) were appropria
te and adequately described.",,"Salmonella/Mammalian-Microsome Pre
-Incubation Mutagenesis Assay (Study #009-617-278-2). EG&G Mason R
esearch Institute, Rockville, Maryland, July 18, 1979 (M260-79).",
"Y"
15022002093307.0,2,2/24/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-26-79
Purity: 95.6%
Additions: None reported
Solvent Carrier: Ethanol
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.5300","Mouse lymph
oma assay","Non-bacterial","Unknown",1979,"L5178Y Mouse Lymphoma c
ells","9,000 x g supernatant (4 ml) prepared from adult rat livers
induced by Aroclor","0.0042-0.056 ul/ml (w/o activation) and 0.00
75-0.1 ul/ml (with activation)","Not referenced.","The purpose of
this study was to evaluate MCTR-26-79 for specific locus forward m
utation induction in the L5178Y thymidine kinase (TK) mouse lympho
ma cell assay (Clive, D. and J.F.S. Spector. 1975. Laboratory pr
ocedure for assessing specific locus mutations at the TK locus in
cultured L5178Y mouse lymphoma cells. Mutation Res. 31: 17-29).

Prior to use in the assay, L5178Y cells which were actively growin
g in culture were cleansed to reduce the frequency of spontaneousl
y occurring TK-/- cells. One ml of THMG stock solution was added
to a 100 ml cell suspension containing 0.3E+6 cells/ml. The cultu
re was gassed with 5% carbon dioxide in air and placed on a enviro
nmental incubator shaker at 125 rpm and 37 degrees C. After 24 hr
s, the THMG was removed and the cells rinsed and reinstated in cul
ture at 2E+4 cells/ml. The cells were ready for use after 72 hrs
incubation. The cell populaion density of the prepared cultures w
as determined by adding 1 ml sample of cells to 9 ml of 0.1% tryps
in, incubating at 37 degrees C for 10 minutes, and making three co
unts per sample with the Coulter Counter. Based on the determinat
ion of the number of cells/ml, a 300 ml cell suspension containing
1E+6 cells/ml was prepared, and 6 ml aliquots were dispensed in 4
4 Corning polypropylene centrifuge tubes.

In order to determine the optimal dose levels of the test substanc
e, a preliminary toxicity test with and without S-9 activation was
conducted. The test substance was dissolved in ethanol and conce
ntrations ranged from 0.0001 to 10 ul/ml. Based on the toxicity t
est, the test substance concentrations (w/o activation) were 0.004
2, 0.0056, 0.0075, 0.010, 0.013, 0.018, 0.024, 0.032, 0.042, and 0
.056 ul/ml. Test substance concentrations (with activation) were
0.0075, 0.010, 0.013, 0.018, 0.024, 0.032, 0.042, 0.056, 0.075, an

d 0.1 ul/ml. The test substance was added to each appropriately l abeled tube in amounts at which the final solvent concentration wa s nontoxic to the cell suspension. Either 4 ml of S-9 activation mixture or 4 ml of medium was added to the tubes to yield a final cell suspension of 0.6E+6 cells/ml.

To establish the background level of TK-/- colonies, two control t ubes received solvent only. Two concentrations of ethylmethanesul fonate (EMS; 1.0 and 0.5 ul/ml) and 7,12-dimethylbenz(a)anthracene (7,12-DMBA; 7.5 and 5.0 ug/ml) were used as positive controls for direct acting mutagens and promutagens, respectively. All tubes were gassed with 5% carbon dioxide in air and placed on the Bellco roller drum apparatus for 4 hrs at 37 degrees C. The preparation and addition of the test substance was carried out under amber li ghting and the cells were incubated in the dark during the 4-hr ex posure period. At the end of the exposure period, the cells were washed twice and resuspended in 20 ml of medium, gassed with 5% ca rbon dioxide in air, and replaced on the roller drum apparatus at 37 degrees C.

Expression Time: In order for induced mutations to be expressed, t he cells must undergo several divisions. After the initial exposu re to the test substance, the cells were incubated for 3 days with a cell population adjustment every 24 hrs. The adjustment was ma de by taking daily cell counts and then replacing a volume of cell s with fresh medium which yielded a cell population density of 0.3 E+6 cells/ml.

Cloning: At the end of the expression period, the cells were place d in a restrictive medium (cloning medium with either BUdR, 50 ug/ ml, or Trifluorothymidine, 1 ug/ml) to allow the TK-/- cells to gr ow. The cloning medium contained 0.32% Noble agar which maintaine d the cells in suspension and allowed them to form discrete coloni es of TK-/- cells.

General Preparation:For cloning, the test substance dose levels wh ich exhibited toxicity from 10% to 90% growth inhibition during th e expression period were selected. Two Florence flasks per concen tration to be cloned and two per control tubes were labeled with t he test substance concentrations and whether or not they were acti vated. For each pair of flasks, one was labeled R.M. (restrictive medium) and one was labeled V.C. (viable count). Each flask was prewarmed to 37 degrees C, filled with 100 ml of C.M., and placed on the shaker at 37 degrees C until use.

Six 100 mm petri plates per test substance concentration were labe led with the concentration, whether or not activation was used, an d experiment number. Three of the six were labeled R.M. and three were labeled V.C.

Cell Plating: Cell counts were made for each tube to determine the volume of each cell population which yielded 3E+6 cells. This vo lume was removed, the remainder of the cells were discarded, and t he 3E+6 cells were replaced in the centrifuge tube. The cells wer e centrifuged at 500 x g for 10 minutes, and the supernatant, exce pt for 2 ml, was removed. The cells were resuspended in the remai ning 2 ml of medium and placed in the R.M. flask labeled with the corresponding test substance concentration.

A 5E-4 dilution was carried out by adding 1.0 ml of the R.M. flask suspension to a test tube containing 9 ml of medium, adding 1.0 m l of this to 4 ml of medium, and adding 1.0 ml of that dilution to the appropriate V.C. flask containing 100 ml of C.M. After the d ilution, 1 ml of stock solution of the restrictive agent (BUdR or TFT) was added to the R.M. flask, and both the R.M. flask and the V.C. flask were placed on the shaker at 125 rpm and 37 degrees C.

After 15 minutes, the flasks were removed one at a time, and 33 ml of the cell suspension was pipetted into each of three appropriat ely labeled petri plates. To accelerate the gelling process, the plates were placed in cold storage (4 degrees C) for 20 minutes. The plates were removed and incubated at 37 degrees C in a humidif ied 5% carbon dioxide atmosphere for 10 days.

After the 10-day incubation period, both the R.M. plates and V.C. plates were scored for the total number of colonies per plate. Th ree counts per plate were made on a New Brunswick Biotran II Autom ated Colony Counter. The mutation frequency was determined by div iding the average number of colonies in the three R.M. plates by t he average number of colonies x 1E+4 in the three corresponding V. C. plates. By comparing the mutation frequency of three treated p lates to that of the control plates, the presence of a significant level of mutagenic activity can be detected.","Negative","0.1 ul/ ml with and without activation","With metabolic activation","There were no significant differences noted (p value not given).","The initial toxicity test conducted on the test substance with and wit hout activation indicated respective threshold levels of complete toxicity of 0.1 ul/ml. Based on this data, the compound was teste d in the assay over a range of concentrations from 0.0018 to 0.1 u l/ml both with and without activat
ion.

After the three day expression period, ten cultures without activa tion and ten cultures with activation were selected for cloning ba sed on their degree of toxicity. The nonactivated cultures were c loned at 0.056, 0.042, 0.032, 0.024, 0.018, 0.013, 0.010, 0.0075, 0.0056, and 0.0042 ul/ml. The cultures receiving metabolic activa tion were cloned at 0.1, 0.075, 0.056, 0.042, 0.032, 0.024, 0.018, 0.013, 0.010, and 0.0075 ul/ml.

The mutant frequency (per 1E+4 surviving cells) for nonactivation cultures (see above for concentrations) was 0.7, 0.8, 0.9, 0.6, 1. 0, culture lost, 1.0, 1.0, 0.8, and 0.7, respectively. The solven t controls averaged 0.7. The mutant frequency for activation cult ures was 0.8, 0.7, 0.9, 1.0, 1.0, 0.8, 0.9, 0.7, 0.8, and 0.6. Th e solvent controls averaged 0.7.

The total compound toxicity data (% growth) for the nonactivated c ultures was 51, 89, 68, 91, 111, 84, 97, 101, 89, and 88%. The so lvent controls averaged 125%. The total compound toxicity data fo r the activated cultures was 24%, 83%, 87%, 84%, 72%, 79%, 82%, 91 %, 84%, and 93%. The solvent controls averaged 146%. Positive co ntrols produced mutant frequencies of 28.1% (EMS, 1.0 ul/ml) and 4 .2% (7,12-DMBA, 5.0 ug/ml).

The results of the Cloning Data for both nonactivated and the acti vated cultures indicate that all of the test cultures which were c loned exhibited mutant frequencies that were not significantly dif ferent from the solvent control cultures. The results from the To xicity Data indicate that the nonactivated cultures exhibited tota l growth over a range of 51 to 111%, and the activated cultures ex hibited total growth over a range of 24 to 93%.","The results indi cate that under the test conditions, MCTR-26-79 did not cause a si gnificant increase in the mutant frequency of any of the test conc entrations in the presence or absence of S-9 activation. However, the narrow concentration range which produced from 0 to 100% toxi city (0.01 and 0.1 ul/ml, respectively) limited the number of sig nificant data points on which to base this judgement.","Acceptable ","The key parameters (i.e., concentrations, use of positive contr ols) were appropriate and adequately described.",,"Evaluation of c ompound MCTR-26-79 (MRI #278) for
mutagenic potential employing the L5178Y TK +/- mutagenesis assay. Study No.: 009-617-278-7. EG&G Mason Research Institute, Rockvi lle, Maryland, June 6, 1979 (M261-79).","Y"
15022002093307.0,3,2/26/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-26-79
Purity: 95.6%
Additions: None reported
Solvent Carrier: Dimethylsulfoxide (DMSO)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.5500","DNA damage and repair assay","Bacterial","Yes",1979,"Escherichia coli and Sal monella typhimurium","Adult male Sprague-Dawley rat liver microsom es induced by Aroclor 1254 (0.5 ml)","0.005 to 6.0 ul/plate","Not

referenced.","The purpose of this study was to evaluate the abilit y of the test substance to react with cellular DNA using DNA Damag e/Repair Assay. E. coli strains WP2uvrA+recA+ and WP100uvrA-recA- and Salmonella typhimurium strains TA1978uvrB+ and TA1538uvrB- ar e used in the DNA Repair assay.

Top agar was initially prepared with 8 g/l Difco Bacto Agar and 5 g/l NaCl. After autoclaving, the molten agar was distributed in 1 00 ml aliquots into sterile bottles where it was stored at room te mperature. Immediately before its use in the DNA repair assay, th e top agar was melted and supplemented with 10 ml/100 ml agar of a sterile solution of 10XSA (containing 5.0 mM L-histidine and 0.5 mM biotin) for plating Salmonella or 10XSC (containing 5.0 mM L-tr yptophan) when plating E. coli. The top agar was also supplemented with 6 ml of Nutrient Broth and 19 ml of deionized, distilled wat er per 100 ml top agar. Bottom agar was the Vogel-Bonner minimal medium E described by Ames. Dimethylsulfoxide (DMSO) was the solv ent used in the assays.

All tester strains were stored in liquid nitrogen, and fresh cultu res were inoculated directly from these frozen stocks. Broth cult ures were grown overnight at 37 degrees C with shaking. Approxima tely two hours before exposure to the test substance, samples of t he overnight cultures were appropriately diluted into fresh broth with continued shaking at 37 degrees C. The resulting log phase c ultures were then diluted in broth to a final cell concentration o f 1E+5 cells/ml.

Diluted tester strains (0.1 ml) were added to an appropriate volum e of test substance. S-9 mix (adult male Sprague-Dawley rat liver microsomes induced by Aroclor 1254, 0.5 ml) or Sham S-9 mix (0.5 ml) was added to the appropriate tubes. A final volume of 1 ml co ntained in a 13 mm tube was used for incubation. Each strain was incubated with the solvent or test substance for 90 minutes with s haking at room temperature. At the end of the incubation period, 100 ul aliquots of the incubation mixture were added to 2.5 ml of appropriately supplemented top agar and plated on Vogel Bonner bot tom agar in triplicate to determine viable counts. The plates wer e incubated for 48 hrs at 37 degrees C. Positive controls were 2-aminofluorene (200 ug/ml, activation) and 4-nitroquinoline-1-oxide (0.075 ug/ml, nonactivation). The negative control was penicilli n (15 ug/ml).

All colonies were counted with a BioTrans II automated colony coun ter whenever possible. Colony counts were made by hand when autom ated counting was not possible. Data from replicate platings were averaged and a Survival Index was calculated for each pair of tes ter strains. Each tester strain was exposed to four doses of the test substance.

The percent survival of each tester strain was calculated by compa ring the number of treated survivors to the solvent treated surviv ors. The Survival Index was then determined by dividing the perce nt survival of the repair deficient strain with that of the repair proficient strain. For data comparison purposes, any repair defi cient strain colony with an average equal to zero was treated as a colony of one. This allowed for generation of a maximum survival index. Decreasing survival indices with increasing concentration s of test substance were considered indicative of genotoxicity in this assay.","Ambiguous","Variable depending on condition and stra in.","With metabolic activation","Not reported.","Inherent test sy stem variabilities necessitated repeated testing of MCTR-26-79. T hese variabilities include: 1. Strong preferential kill of the re pair proficient strains by the negative control. 2. Strong prefer ential kill of the repair proficient strains by the test article. 3. Lack of preferential kill of t
he repair deficient strains by the positive controls, especially w ith activation. 4. Unexplained reduction of plating efficiency of the repair deficient strains. 5. Narrow range of test substance concentration which demonstrated acceptable toxicity. The repeat ed testing of MCTR-26-79 demonstrated preferential kill of the rep air deficient strains. However, no consistent pattern of preferent ial kill was evident.

A description of the experimental results is as follows. Date: 7/ 3/79 (nonactivation, 0.005, 0.012, 0.018, and 0.030 ul/ml). E. col i: Negative control - Heavy preferential kill of repair proficient strain; Positive control - OK; Treated plates - No signficant tox icity demonstrated on either strain. Salmonella: Negative control - Slight preferential kill of repair deficient strain; Positive co ntrol - OK; Treated plates - No significant toxicity demonstrated on either strain.

Date: 7/6/79 (activation, 0.005, 0.012, 0.018, and 0.030 ul/ml). E . coli: Negative control - Slight preferential kill of repair prof icient strain; Positive control - OK; Treated plates - No signific ant toxicity demonstrated on either strain. Salmonella: Negative c ontrol - Some preferential kill of repair deficient strain; Positi ve control - OK; Treated plates - No significant toxicity demonstr ated on either strain.

Date: 7/11/79 (nonactivation, 0.03, 0.3, 3, and 6 ul/ml). E. coli: Negative control - Some preferential kill of repair proficient st rain; Positive control - OK; Treated plates - Total toxicity at 0. 3 ul and above. Salmonella: Negative control - Low counts of solve nt control; Positive control - OK; Treated plates - Total toxicity at 0.3 ul and above.

Date: 7/12/79 (activation, 0.03, 0.3, 3, and 6 ul/ml). E. coli: Ne gative control - Heavy preferential kill of repair proficient stra in; Positive control - OK; Treated plates - Total toxicity at 0.3 ul and above. Salmonella: Negative control - Preferential kill of repair deficient strain; Positive control - OK, but no greater tha n negative control; Treated plates - Total toxicity at 0.3 ul and above.

Date: 8/30/79 (nonactivation, 0.3, 0.6, 1.2, and 2.4 ul/ml). E. co li: Negative control - Preferential kill of repair proficient stra in; Positive control - OK; Treated plates - Total toxicity. Salmon ella: Negative control - Some preferential kill of repair proficie nt strain; Positive control - OK; Treated plates - Total toxicity.

Date: 8/31/79 (activation, 0.3, 0.6, 1.2, and 2.4 ul/ml). E. coli: Negative control - Heavy preferential kill of repair proficient s train; Positive control - OK; Treated plates - Total toxicity. Sal monella: Negative control - Reasonable toxicity; Positive control - OK; Treated plates - Total toxicity.

Date: 9/6/79 (nonactivation, 0.03, 0.1, 0.2, and 0.3 ul/ml). E. co li: Negative control - Insufficient growth of WP100; Positive cont rol - Not meaningful due to insufficient growth of WP100; Treated plates - Insufficient growth of WP100. Salmonella: Negative contro l - Insufficient growth of TA1538; Positive control - Not meaningf ul due to insufficient growth of TA1538; Treated plates - Preferen tial kill of repair proficient strain at 0.03 ul. Total toxicity at all higher doses.

Date: 9/12/79 (activation, 0.02, 0.03, 0.05, and 0.1 ul/ml). E. co li: Negative control - Preferential kill of repair proficient stra in; Positive control - OK; Treated plates - Some preferential kill of repair proficient strain. Salmonella: Negative control - Some preferential killing of repair proficient strain; Positive control - Preferential kill of repair proficient strain; Treated plates - Preferential kill of repair proficient strain.

Date: 9/27/79 (nonactivation, 0.05, 0.15, 0.25, and 0.35 ul/ml). E . coli: Negative control - Insufficient growth; Positive control - Not meaningful due to insufficient growth of WP100; Treated plate s - Insufficient growth of tester strains. Salmonella: Negative co ntrol - Insufficient growth of TA1538; Positive control - OK; Trea ted plates - Dose range too high.

Date: 9/27/79 (activation, 0.05, 0.15, 0.25, and 0.35 ul/ml). E. c oli: Negative control - Insufficient growth of tester strains; Pos itive control - OK; Treated plates - Inconclusive data. Salmonella : Negative control - Insufficient growth of TA1538; Positive contr ol - Preferential kill of repair proficient strain; Treated plates - Preferential kill of repair proficient strain.

Date: 10/16/79 (nonactivation, 0.025, 0.05, 0.075, and 0.1 ul/ml). E. coli: Negative control - Heavy preferential kill of repair pro ficient strain; Positive control - OK; Treated plates - Heavy pref erential kill of repair proficient strain. Good toxicity range ove rall. Salmonella: Negative control - Insufficient growth of TA153 8; Positive control - OK; Treated plates - Preferential kill of re pair proficient strain at three dose levels. Preferential kill of repair proficient strain at one dose level. Data inconsistent. Good toxicity range overall.

Date: 10/17/79 (activation, 0.025, 0.05, 0.075, and 0.1 ul/ml). E. coli: Negative control - Heavy preferential kill of repair profic ient strain; Positive control - OK; Treated plates - Preferential kill of repair proficient strain. Overall colony counts much too h igh for good counting accuracy. Salmonella: Negative control - So me preferential kill of repair proficient strain; Positive control - OK; Treated plates - Preferential kill of repair proficient str ain.

Date: 11/16/79 (nonactivation, 0.02, 0.06, 0.09, and 0.15 ul/ml). E. coli: Negative control -Preferential kill of repair proficient strain; Positive control - OK; Treated plates - Preferential kill of repair proficient strain at two dose levels and preferential ki ll of repair deficient strain at one dose level. Salmonella: Nega tive control - Preferential kill of repair proficient strain; Posi tive control - OK; Treated plates - Preferential kill of repair pr oficient strain at one dose level and then preferential kill of th e repair deficient strain at the next dose level.

Date: 11/16/79 (activation, 0.02, 0.06, 0.09, and 0.15 ul/ml). E. coli: Negative control -Preferential kill of repair proficient str ain; Positive control - OK; Treated plates - Slight preferential k ill of repair proficient strain at top dose level. Salmonella: Ne gative control - OK; Positive control - No significant preferentia l kill of repair deficient strain; Treated plates - Preferential k ill of repair proficient strain at top dose level.

Date: 11/20/79 (activation, 0.02, 0.06, 0.09, and 0.15 ul/ml). E. coli: Negative control - Data not available; Positive control - Da ta not available; Treated plates - Data not available. Salmonella : Negative control - Preferential kill of repair proficient strain ; Positive control - Weaker than usual preferential kill of repair deficient strain; Treated plates - Inconsistent preferential kill of tester strains.

Date: 11/27/79 (activation, 0.02, 0.06, 0.09, and 0.15 ul/ml). E. coli: Negative control -Insufficient growth of WP100; Positive con trol - Insufficient growth of WP100; Treated plates - Insufficient growth of WP100. Salmonella: Negative control - Some preferentia l kill of repair proficient strain; Positive control - Weaker than usual preferential kill of repair deficient strain; Treated plate s - Preferential kill of repair deficient strain at top two dose l evels.

Date: 11/27/79 (nonactivation, 0.02, 0.06, 0.09, and 0.15 ul/ml). E. coli: Negative control -Insufficient growth of WP100; Positive control - Insufficient growth of WP100; Treated plates - Insuffici ent growth of WP100. Salmonella: Negative control - Preferential kill of repair proficient strain; Positive control - OK; Treated p lates - Inconsistent preferential kill of tester strains.

Date: 12/11/79 (activation, 0.02, 0.06, 0.09, and 0.15 ul/ml). E. coli: Negative control -Preferential kill of repair proficient str ain; Positive control - OK; Treated plates - Clear preferential ki ll of repair proficient strain at top dose. Salmonella: Negative control - Some preferential kill of repair proficient strain; Posi tive control - OK; Treated plates - Some preferential kill of repa ir proficient strain at first three dose levels and then preferent ial kill of repair deficient strain at top dose level.

Date: 12/11/79 (nonactivation, 0.02, 0.06, 0.09, and 0.15 ul/ml). E. coli: Negative control -Preferential kill of repair proficient strain; Positive control - OK; Treated plates - Inconsistent prefe rential kill of repair proficient strain. Salmonella: Negative co ntrol - Some preferential kill of repair proficient strain; Positi ve control - OK; Treated plates - Preferential kill of repair prof icient strain.","The results of the Bacterial DNA Damage/Repair Su spension Assay indicate that MCTR-26-79 did cause an inconsistent pattern of preferential killing of the repair deficient strains. However, due to the inherent variability of this test system and t he lack of a significant comparative data base, the results of thi s study should be evaluated only in conjuction with other in vitro tests that monitor genetic activity. Uncorroborated results from this test system should not be heavily weighted.","Unacceptable", "The data is not reliable based on the variability as described ab ove.",,"Bacterial DNA Damage/Repai
r Suspension Assay. Study No.: 009-617-278-6; MCTR-26-79. EG&G M ason Research Institute, Rockville, Maryland, February 7, 1980 (M2 63-79).","Y"
15022002093307.0,5,3/1/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: TACU #01038003

Purity: Assume 100% for dosing calculations
Additions: None reported
Solvent Carrier: Dimethylsulfoxide (DMSO, 50 ul per 3 ml)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.5575","Mitotic rec ombination in Saccharomyces cerevisiae","Non-bacterial","Yes",1982 ,"Saccharomyces cerevisiae","A 9,000 x g supernatant (1.5 ml) prep ared from adult rat liver induced by Aroclor","0.020, 0.039, 0.078 , 0.156, and 0.312 ul per 3 ml","Not referenced.","The objective o f this study was to evaluate the test substance for genetic activi ty in the yeast strain Saccharomyces cerevisiae D5 with and withou t metabolic activation. Stocks of yeast strain D5 were maintained as single colony isolates at 4 degrees C on plates of yeast compl ete medium. Working stock suspensions of the strain were obtained from overnight cultures. A single colony isolate was suspended i n yeast complete broth and incubated at 30 degrees C for 16-18 hrs . An aliquot from this culture was used in the assays.

Doses used in these assays were selected from a preliminary toxici ty test performed on the strain D5. For the preliminary toxicity test, 14 doses from 0.02 to 150 ul per 3 ml were used with station ary phase cultures. As a result of this test, the mitotic recombi nation assays were conducted at doses of 1, 5, 10, 25, and 50 ul p er 3 ml using logarithmically growing cells. The test substance e xhibited complete toxicity at all the doses employed. As a result , a second cytotoxicity test was conducted using doses ranging fro m 0.039 to 5.0 ul per 3 ml using logarithmically growing cells.

The toxicity studies were conducted as follows: to a sterile 20 ml scintillation vial, 0.05 to 0.15 ml of a solution of the test sub stance was added to give the appropriate dose. The indicator orga nism was then added (0.3 ml; approximately 1E+8 cells/ml) along wi th 2.55 to 2.65 ml of 0.2 M phosphate buffer, pH 7.4. The above m ixture was incubated for 3 hrs at 30 degrees C in a rotary shaker. After incubation, cell survival was determined by adding 0.25 ml of 1E-4 dilution of the suspension to 2 ml of molten (45 degrees C) overlay agar, which was poured onto the respective yeast comple te plates. These plates were incubated for three days and scored.

The procedure used for the recombination assay was based on the me thod of Zimmermann (Mutation Res., 21:263-269, 1973). Tests were c onducted in sterile 20 ml scintillation vials. To sterile vials t he following were added: 0.05 to 0.15 ml of a solution of the test chemical to give the appropriate dose, 0.3 ml of the indicator or ganism (approximately 1E+8 cells/ml), and 2.55 to 2.65 ml of 0.2 M phosphate buffer, pH. 7.4. This mixture was incubated at 30 degr ees C on a rotary shaker for approximately 3 hrs. Samples were th en removed, diluted in 0.15 M saline and plated onto yeast complet e medium. All plates were incubated at 30 degrees C for approxima tely four days. The plates were then refrigerated 1-3 days to int ensify the color of the pigmented colonies. The plates were scree ned for pigmented colonies and sectors using a dissecting microsco pe with variable magnification.

The activation assay was run concurrently with the nonactivation a ssay. The only difference was the addition of 1.05 to 1.15 ml of 0.2 M phosphate buffer, pH 7.4, and 1.5 ml of S9 mix (a 9,000 x g supernatant prepared from adult rat liver induced by Aroclor) to t he sterile vials.

A negative control consisting of the solvent dimethylsulfoxide (DM SO, 50 ul per 3 ml) was used. The positive control for the nonact ivation assay was ethylmethanesulfonate (EMS, 1%). Sterigmatocyst in (5 ug/ml) was used as the positive control for the activation a ssay.

The phenotypic expression (or the events) of reciprocal recombinat ion in the yeast strain are the Red-Pink cells representing the ho mozygocity of the two recessive alleles. The nonreciprocal recomb ination which is also known as gene conversion is again a non-muta tional genetic event and can occur in dividing or resting cells. At the two-strand stage of nondividing cells of this diploid strai n, the nonreciprocal recombination forms red or pink colonies and at the four-strand replicative stage during cell division, gene co nversion brings about red-white sectored colonies. The results of the assays will be considered positive if the total events in a t est are equal to or greater than 2 times the spontaneous events. An accompanying dose-related effect is also necessary to give conf idence to the increase.","Negative","Complete toxicity at 0.625 ul and higher (without activation)","With metabolic activation","The test substance did not induce mitotic recombinations.","Initially , a cytotoxicity test was performe
d on the test substance at 14 doses ranging from 0.02 to 150 ul us ing the stationary phase cultures of the yeast strain. Complete t oxicity was not observed at any dose. As a result, the mitotic re combination assays were conducted at 1, 5, 10, 25, and 50 ul per 3 ml using logarithmically growing cells. However, the test substa nce exhibited complete toxicity at all doses. A second cytotoxici ty test was conducted on the test substance at 0 (solvent control) , 0.039, 0.078, 0.156, 0.313, 0.625, 1.25, 2.5, and 5.0 ul per 3 m l. The percent survival relative to control was 100, 107, 96, 39, 3, 0, 0, 0, and 0%, respectively. Based on these results, the mi totic recombination assays were repeated at 0.020, 0.039, 0.078, 0 .156, and 0.312 ul per 3 ml with activation and nonactivation. In the nonactivation assay, the test substance produced mitotic reco mbinations at approximately the same percent as the solvent contro

ls at all doses except the 0.312 ul dose. At this dose the percen
t aberrant colonies were 2.19 but
the absolute number of abberrant colonies was less than the solven
t controls. Therefore, this increase was not considered significa
nt. In the activation assay, the test substance produced mitotic
recombinants at the same percent or lower than the solvent control
s. The results for the nonactivation assays were as follows (resu
lts for solvent control, positive control, 0.020, 0.039, 0.078, 0.
156, and 0.312 ul per 3 ml, respectively): Total Aberrant Colonies
; 6, 97, 2, 7, 4, 3, and 3. Total Number of Colonies Scored; 1526
, 1703, 1796, 1567, 1486, 1527, and 137. Percent Aberrant Colonie
s; 0.39, 5.7, 0.11, 0.45, 0.27, 0.20, and 2.19. Cfu per ml (colon
y forming units); 1.53E+7, 1.7E+7, 1.8E+7, 1.6E+7, 1.5E+7, 1.53E+7
, and 1.4E+6. The results for the activation assays were as follo
ws (results for solvent control, positive control, 0.020, 0.039, 0
.078, 0.156, and 0.312 ul per 3 ml, respectively): Total Aberrant
Colonies; 3, 31, 3, 5, 0, 3, and 1. Total Number of Colonies Scor
ed; 1436, 702, 1638, 1775, 1647, 1
641, and 1770. Percent Aberrant Colonies; 0.21, 4.42, 0.18, 0.28,
0.0, 0.18, and 0.06. Cfu per ml (colony forming units); 1.44E+7,
7.0E+6, 1.64E+7, 1.8E+7, 1.65E+7, 1.64E+7, and 1.8E+7.","The test
substance, TACU #01038003 did not induce mitotic recombinations i
n any of the assays conducted in this evaluation and was considere
d genetically inactive to the indicator strain Saccharomyces cerev
isiae strain D5.","Acceptable","The key parameters (i.e., doses, u
se of positive and negative controls, etc.) were appropriate and a
dequately described in the study.",,"Mutagenicity Evaluation of Pa
ra Ethyltoluene (TACU #01038003, Study Number 20733) in the Mitoti
c Recombination Assay with the Yeast Strain D5. LBI Project No.:
20988, Litton Bionetics, Kensington, Maryland, October, 1982 (733-
82).","Y"
15022002093307.0,4,2/26/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: MCTR-26-79
Purity: 95.6%
Additions: None reported
Solvent Carrier: Acetone (5 ul/ml)
Contaminants: None reported
Chemical formula: C9H12",,"EPA OPPTS Method 870.8800","Cell Transf
ormation","Non-bacterial","Yes",1979,"C3H/10T1/2 CL8 mouse embryo
culture","None","0.039, 0.078, 0.156, and 0.3125 ug/ml","None refe
renced.","The purpose of this study was to evaluate the carcinogen
ic potential of the test substance using the C3H/10T1/2 cell trans
formation assay. The C3H/10T1/2 CL8 cells used in this study were
derived from the cryopreserved Lot No. 3-2-1978, Passage No. 8.

A dose range study to determine the toxicity of the test substance
was conducted prior to testing for transformation potential. The
test substance was tested in duplicate using 60 mm culture plates

seeded with 200 cells per plate and grown in 5 ml of BME medium s
upplemented with 10% fetal bovine serum. The test substance was t
ested using 14 2-fold dilutions over a concentration range of 10 u
l/ml to 0.0013 ul/ml. After 18 hrs, the cells were refed with fre
sh medium and incubated for 8 days. The plates were washed with P
BS, fixed with absolute methanol and stained with Giemsa stain. T
he number of colonies per plate was counted and the cloning effici
ency (CE) and the relative cloning efficiency (RCE) were determine
d by the following formula: CE=average number of colonies/plate di
vided by the number of cells seeded/plate x 100. RCE=test culture
cloning efficiency divided by solvent control cloning efficiency
x 100.

The transformation potential of the test substance was tested at f
our dose levels in decreasing 2-fold dilutions from the concentrat
ion which caused 74% reduction in cloning efficiency. Twelve repl
icate plates seeded with 1000 cells/plate were treated by 0.039, 0
.078, 0.156, and 0.3125 ul/ml. The positive control (7,12-dimethy
lbenz[a]anthracene) was tested at 0.5 ug/ml and 0.25 ug/ml. In pa
rallel with the test plates, four toxicity plates containing 200 c
ells each were treated with the sample compound dilutions. Approx
imately 18 hrs after treatment, the test substance was removed fro
m all assay and toxicity plates which were then refed with growth
medium and reincubated. The toxicity plates were incubated for 8
days, stained and the relative cloning efficiency was determined a
s previously described. This was to assure that the assay was bei
ng conducted at compound concentrations approaching the LD50. The
remaining culture plates were refed weekly with BME medium supple
mented with 5% fetal bovine serum.
At 35 days after removal of the test substance, all plate culture
s were washed, fixed, stained, examined microscopically and macros
copically, and scored for transformation.

Focal areas of transformation were classified according to the cri
teria of Reznikoff (Cancer Research 33: 3239-3249, 1973) as follo
ws: Type I. Foci composed of monolayer cells are more densely pa
cked than the background cells. This type is not considered malig
nant and was not scored. Type II. Foci show massive piling up in
to virtually opaque multilayers. The cells are only moderately po
lar, thus criss-crossing is not pronounced. Type III. Foci are c
omposed of highly polar, fibroblastic, multilayered, criss-crossed
arrays of densely stained cells.","Negative","26% relative clonin
g efficiency at 0.312 ul/ml (without activation)","Without metabol
ic activation","Not reported.","In the initial toxicity test, the
test substance at concentrations of solvent control, 0.0013, 0.002
5, 0.0049, 0.0098, 0.0195, 0.059, 0.078, 0.156, 0.312, 0.625, 1.25
, 2.5, 5, and 10 ul/ml showed a relative cloning efficiency of 100
, 99, 95, 99, 88, 62, 48, 52, 39, 26, 0, 0, 0, 9, and 0%, respecti

vely.

The toxicity study conducted in parallel to the transformation ass ay showed the reduction in cloning efficiency for solvent, DMBA (0 .5 ug/ml), DMBA (0.25 ug/ml), and test substance concentration of 0.039, 0.078, 0.156, and 0.3125 ul/ml to be 100, 51, 70, 98, 91, 7 8, and 42%, respectively.

In the transformation assay, the solvent control plates showed thr ee Type II foci. The test substance plates showed one Type II foc us at 0.156 ul/ml, two Type II foci at 0.078 ul/ml and three Type II foci at 0.039 ul/ml. None of the solvent control plates or tes t substance plates showed Type III foci. The overall transformati on frequency in test substance treated plates was less than in the solvent control plates. Both solvent control and test substance plates showed a large number of Type I foci composed of cells that were more densely packed than the background cells. However, the frequency of Type I foci observed in the test substance plates wa s greater than in the solvent control plates. The positive contro l (DMBA) at 0.5 ug/ml and 0.25 ug/ml showed extensive development of Type III and Type II foci.","Test substance MCTR-26-79 was test ed in the C3H/10T1/2 Cell Transformation Assay at four dose levels ranging from 0.039 to 0.3125 ul/ml. In this study, the solvent c ontrol showed a low background of
Type II transformed foci. However, the frequency of Type II foci in the test substance plates was less than in the solvent control. The results of the assay, therefore, indicate that the test subs tance did not cause morphological transformation of cells in the C 3H/10T1/2 Cell Transformation Assay.","Acceptable","All key parame ters (i.e., dose selection, postive controls, etc.) were appropria te and adequately described in the study.",,"An Evaluation of Carc inogenic Potential of MCTR-26-79 Employing the C3H/10T1/2 Cell Tra nsformation Assay. Study No.: 009-617-278-8. EG&G Mason Research Institute, Rockville, Maryland, September 26, 1979 (M264-79).","Y "

"DSN","TestNo","Rev_Date","TestSubstRem","ChemCat","Method","GLP","Year","MethodRem","Prec","BoilVal","Upper","Unit","Pressure","PresUnit","Decomposition","ResultsRem","ConcludingRem","Reliability","ReliRem","GeneralRem","RefRem","Completed"
15022002093307.0,1,3/18/02 0:00:00,"Toluene, p-ethyl-
Test Article ID#: Toluene, p-ethyl-
Purity: Assume 100%
Additions: Unknown
Solvent Carrier: Unknown
Contaminants: Unknown
Chemical formula: C9H12",,"Unknown","Unknown",,"Unknown","=",162,0,"§C",760.00,"mm Hg",,"Report not evaluated.","Report not obtained and evaluated.","Unknown","The report was not evaluated.",,"Standard MSDS.","Y"